

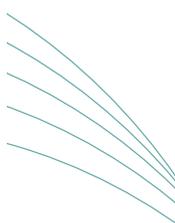
# Impacts of plant protection products on biodiversity and ecosystem services

S. Leenhardt, L. Mamy, S. Pesce, W. Sanchez, eds

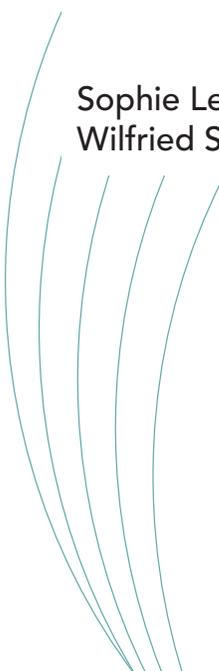




# Impacts of plant protection products on biodiversity and ecosystem services



Sophie Leenhardt, Laure Mamy, Stéphane Pesce,  
Wilfried Sanchez, editors



Éditions Quæ



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This book is a synthesis of a collective scientific assessment (CSA) requested jointly by the Ministries of Ecology, Agriculture and Research. It was produced by a group of scientific experts without prior approval by either the sponsors, INRAE or Ifremer. The present summary, taken from the extended report, is the sole responsibility of its authors.

The documents associated with this assessment are available on the websites of INRAE ([www.inrae.fr](http://www.inrae.fr)) and Ifremer ([www.ifremer.fr](http://www.ifremer.fr)).

This document is the condensed version of the extended report, authored by the expert committee, *ad hoc* contributors requested by the experts, and DEPE contributors, as listed in the reference below:

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# Foreword

As part of the Ecophyto II+ plan, various expert studies have been conducted in a complementary manner. In June 2021, the French National Institute for Health and Medical Research (Inserm) presented the results of a collective scientific assessment (CSA) on the effects of plant protection products (PPPs) on human health, entitled 'Effects of Pesticides and Health - New Data'. The CSA presented here focuses on the impacts of PPPs on biodiversity and ecosystem services. Another CSA, delivered on 20 October 2022, examines the use of plant diversity in agricultural areas to regulate crop pests.

Requested in March 2020 by the Ministries of the Environment, Agriculture and Research, the present CSA was assigned to INRAE and Ifremer. It updates and supplements previous studies published in 2005 (*Pesticides, agriculture et environnement*) and in 2008 (*Agriculture et biodiversité*).

The results are published on the INRAE and Ifremer websites in three formats. The full 1,408-page extended report provides the background and context of the assessment, describes the method used, contains the full bibliography (more than 4,500 references), provides the scientific framework specific to this CSA, includes all of the analyses conducted by its experts, and presents the general conclusions drawn from them. The summary, which is also the subject of this book, brings together the main findings of the CSA extended report, without citing the entire body of literature used. In this document, references are only cited when the data or examples mentioned are taken directly from a publication. The 14-page summary presents the main conclusions drawn from this collective assessment.

# Introduction

Each year, between 55,000 and 70,000 tonnes of plant protection product (PPP) active ingredients, including those that can be used in organic farming and biocontrol, are sold in France and its overseas territories<sup>1</sup>. These substances are mainly intended for crop protection, with an estimated 2-5% of the total used for non-agricultural practices (i.e. maintenance of gardens, green spaces and infrastructures). They are used in the composition of commercial products incorporating co-formulants that may be associated with adjuvant. After use, they may undergo various biotic and abiotic degradation processes leading to the appearance of transformation products. Crop protection is largely based on synthetic organic molecules and mineral substances, but it can also use biocontrol products, i.e. natural substances from plants, animals or minerals, microorganisms, macroorganisms and semiochemicals (e.g. pheromones, kairomones) that contribute to the control of populations of target organisms. All substances and organisms used for crop protection and maintenance of non-agricultural areas, as well as their co-formulants and adjuvants, are included here under the term 'plant protection products' (PPPs). PPP transformation products are also considered. Although the term 'pesticides' is more widely used in everyday language, PPP has been chosen here to more precisely define the scope of the collective scientific assessment (CSA). This is consistent with the vocabulary used in regulatory documents to distinguish, from pesticides, all biocides used for various purposes, and PPPs used for crop protection or the maintenance of non-agricultural areas (Figure 1). Therefore, the use is what characterises a PPP in relation to other regulatory categories. PPPs are designed to be used directly in the environment, on surfaces that can range from a few dozen square metres to several hundred hectares for a single application. In France, they can potentially cover around 20 million hectares for agricultural treatments<sup>2</sup>, and between 3 and 4 million hectares for non-agricultural areas (Ballet, 2021)<sup>3</sup>. Their use is designed to target organisms that cause damage to crops and beneficial organisms, but they can also cause unintended effects. These include direct effects on the physiology of non-target organisms exposed to PPPs, depending on the environmental fate of these products, as well as indirect effects. The stress on directly impacted organisms has repercussions on the ecological dynamics in which they play roles. This large-scale use, within areas integrated into ecosystems, of molecules intended to eliminate certain

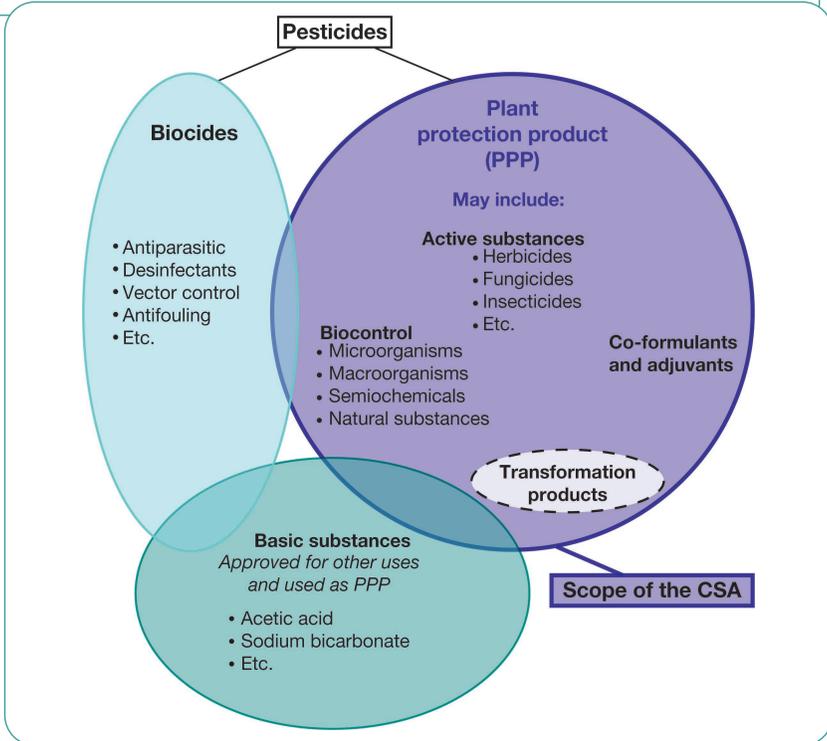
1. Source: Ecophyto monitoring notes: <https://agriculture.gouv.fr/le-plan-ecophyto-quest-ce-que-cest>. Only those French overseas territories falling within the scope of the diffuse pollution fee are included here: Guadeloupe, Martinique, French Guiana, and Réunion.

2. Source: Agreste Statistique agricole annuelle 2020: usable agricultural area (SAU) 28 Mha; surface area under grass (STH) 8 Mha. [https://agreste.agriculture.gouv.fr/agreste-web/disaron/SAANR\\_1/detail/](https://agreste.agriculture.gouv.fr/agreste-web/disaron/SAANR_1/detail/) (accessed 9/01/2023).

3. Teruti survey: stabilised permeable artificial soils (railways, forest tracks, non-agricultural roads, landfills) and other permeable artificial soils (lawns, gardens, parks, roadsides), i.e. about two thirds of the 5 Mha of artificialised land.

organisms considered harmful naturally raises the question of the consequences of their application on biodiversity.

**Figure 1. Range of substances considered (adapted from Pesce *et al.*, 2023a)**



Today's taxonomic and functional biodiversity is the result of evolution. This precious heritage should be preserved first and foremost for its own sake, which does not preclude the use of the resources it offers, but it should be used sustainably and for the common good, as promoted by the International Union for Conservation of Nature (IUCN). It is essential for life and a source of resilience in the context of global changes induced by human activities. In particular, it can help to regulate and limit the imbalances and some of the effects resulting from these global changes. At the same time, these same global changes, through displacement of species' ranges, increases in the amplitude and frequency of extreme events, and changes in the physico-chemical conditions prevailing in the various environments, are weakening biodiversity. When the magnitude of change exceeds the capacity of living organisms to adapt, species disappear or decline, sometimes to the benefit of other species that may become invasive. Habitats and ecosystems are then more or less profoundly modified, as are the associated ecological processes.

Changes in biodiversity under the influence of now clearly identified pressures have been noticeable for many decades. According to the IUCN, 22.7% of the 15,060 European species that have been assessed are threatened with extinction<sup>4</sup>. However, these changes show varying and sometimes contrasting trends depending on the timeframe, geographical areas, species and habitats considered, which makes their characterisation complex. These contrasting trends are evidence of the diverse processes of resilience, adaptation and weakening that coexist. However, it is now clearly established that the erosion of biodiversity is the dominant global trend and that it compromises the capacity of ecosystems to adapt to global change.

The use of PPPs contributes to this dynamic in a paradoxical manner. Although their purpose is to protect crops against species considered harmful, they also contribute to increasing the vulnerability of production by abandoning preventive strategies and/or by stimulating the appearance of harmful species resistant to the PPPs applied, and/or by altering the natural regulatory processes favourable to crops.

Moreover, PPP contamination occurs in addition to that from other chemical substances and other types of pressure, including, for example, the permanent destruction of ecological habitats due to increased urbanisation and the intensification of agricultural and forestry crops. The pressures on biodiversity are therefore multiple and vary greatly depending on the context, including with regard to PPPs. The specific impacts of a substance for a given use on biodiversity as a whole is therefore very difficult to measure quantitatively. However, this question is important from a regulatory perspective for the marketing of products, which can only be marketed if they "have no harmful effects on human or animal health and no unacceptable effects on the environment" (European Commission, 2009b). In the light of this regulatory requirement, numerous alerts have been issued, leading to specific initiatives of various kinds. At the French level, some of these include the National Chlordecone Plan (since 2009), the Glyphosate Exit Plan (2019), the National Strategy on Endocrine Disruptors (since 2014), the referral to the French Agency for Food, Environmental and Occupational Health and Safety (Anses) of SDHIs (succinate dehydrogenase inhibitor fungicides) in 2019, the National Biocontrol Deployment Strategy (2020), and the ban followed by reauthorisation of neonicotinoid insecticides (2021 and 2022). The regulatory assessment of the risk of PPPs to biodiversity is thus subject to conflicting criticisms. On the one hand, it is denounced by some stakeholders as imposing too many constraints on the authorisation and use of PPPs and, on the other hand, it is criticised by others as insufficiently protective of human health and the environment.

## Context

In light of the evidence of the impact of PPPs on the environment (Aubertot *et al.*, 2005b), the first Ecophyto plan was set up in 2008, in conjunction with the adoption by the

4. <https://www.iucnredlist.org/regions/europe> (accessed 9/01/2023).

European Union in 2009 of the Pesticides Package, which is a set of directives and regulations governing the use of PPPs. This public policy framework for PPPs has various components: objectives and action plans for reducing PPP use, rules for assessing and placing PPPs on the market, and mechanisms for monitoring environmental contamination and the resulting unintended effects.

Since 2008, successive versions of the Ecophyto plan have reaffirmed the objective of drastically reducing the use of PPPs and the associated risks. However, the means employed and the actions deployed to this end have not achieved the objectives set, as highlighted in 2019 by the French Court of Auditors<sup>5</sup>.

With regard to the evaluation of products before they are placed on the market, the Pesticides Package and the Ecophyto plan have led to the development of risk indicators, including the specific monitoring of sales of substances considered to be of greatest concern. A campaign to re-evaluate these substances has been initiated, with a view to reducing the range of authorisations and considering their replacement by less dangerous substances. Significant scientific activity has been conducted at EFSA (European Food Safety Authority) at the European level, as well as at Anses at the national level, to improve the methodological framework of the risk assessment process. A revision of the more general framework of this assessment also came into force at the Community level in 2021 following the 2017 citizens' initiative on glyphosate. This includes improvements to transparency (accessibility of studies and data used by the applicant, confidentiality rules, etc.), the opening of EFSA's governance to Member States, parliamentarians and community representatives, and the introduction of a coordinated risk communication plan. These developments have led to the non-renewal or withdrawal of approval for certain substances or uses, while new chemicals have been placed on the market, particularly in the area of biocontrol.

In terms of environmental monitoring, the inclusion of PPPs in monitoring programmes has been progressively strengthened across the various environmental matrices and environments, in line with regulations dedicated to the protection of environments and biodiversity<sup>6</sup>.

## Request for assessment

In this context, axis 2 (research and innovation) of the Ecophyto II+ plan, through its Scientific Steering Committee on 'Research and Innovation' (CSO R&I), proposed in 2019 that a scientific assessment be conducted on 'the effects on biodiversity and alternatives to plant protection products', as a complement to that of Inserm on the effects on human health (Inserm, 2021). On this basis, the Ministries of the Environment, Agriculture

5. Cour des comptes, 2019. Le bilan des plans Écophyto. Référé n° 22109-2659. <https://www.ccomptes.fr/system/files/2020-01/20200204-refere-S2019-2659-bilan-plans-ecophyto.pdf> (accessed 9/01/2023).

6. WFD (Water Framework Directive); Habitats Directive (Directive on the conservation of natural habitats and of wild fauna and flora); Directive on the conservation of wild birds, MSFD (Marine Strategy Framework Directive).

7. <https://agriculture.gouv.fr/le-plan-ecophyto-quest-ce-que-cest> (accessed 9/01/2023).

and Research commissioned two parallel CSAs, one on the impact of PPPs on biodiversity and ecosystem services, and the other on the use of diverse plant cover to regulate pests and protect crops. With regard to the prospects for reducing the use of PPPs, the priority research programme (PPR) '*Cultiver et protéger autrement*' (Growing and Protecting Crops Differently)<sup>8</sup> was also initiated in 2019; its direction is based in part on the foresight study '*Agriculture européenne sans pesticides*' (Pathways to European Pesticide-free Agriculture)<sup>9</sup>, coordinated by INRAE. Finally, this assessment echoes the '*Océan et climat*' (Oceans & Climate) PPR, coordinated since 2021 by Ifremer and the CNRS. One of its themes involves the development of knowledge on the contamination of the marine environment and the effects of this contamination on marine organisms and associated ecosystem services, in order to propose solutions for a clean, healthy, safe and resilient ocean.

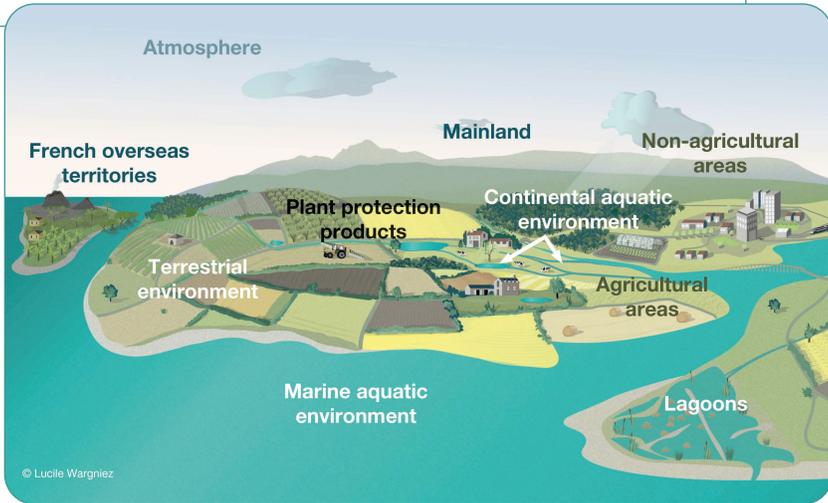
This CSA, on the impacts of PPPs on biodiversity and ecosystem services, also follows on from the 2005 CSA '*Pesticides, agriculture et environnement*' (Pesticides, agriculture and the environment)<sup>10</sup>, which showed that the common use of these substances was leading to environmental degradation and that it was therefore necessary to reduce it. Subsequently, the 2008 CSA '*Agriculture et biodiversité*' (Agriculture and Biodiversity) and the 2017 EFESI study '*l'Évaluation française des écosystèmes et des services écosystémiques*' (French Evaluation of Ecosystems and Ecosystem Services), and in particular its *Assessing Agricultural Ecosystem Services for Better Management* component, demonstrated the complexity of the interrelations between crop protection and biodiversity. Indeed, biodiversity provides essential resources for crops, but it also includes species that are considered harmful to them. Conversely, crop protection treatments targeted at some species have effects on many others, with implications for ecosystem functions and services well beyond the treatment area due to the different modes of transfer of PPPs and their effects. Since the 2005 CSA, crop protection and non-agricultural area management tools have evolved, notably with the banning of certain substances or uses, the introduction of new families of chemicals, and the increasing use of biocontrol treatments. The available data on product use, associated ecotoxicological risks and the state of the environment has also evolved. In particular, the importance of the direct and indirect impacts of PPP use on the functioning of ecosystems is increasingly recognised. In this respect, and given the contextual changes outlined above, a more holistic approach to biodiversity and ecosystem services has been favoured, with a focus on continuums and interdependencies between environments, from the PPP application site to the marine environment. INRAE and Ifremer were therefore jointly tasked with implementing this assessment, given that it considers the entire land-sea environmental continuum. The geographic scope is shown in Figure 2.

8. <https://www6.inrae.fr/cultiver-protoger-autrement/Le-Programme/Presentation> (accessed 9/01/2023).

9. <https://www6.inrae.fr/cultiver-protoger-autrement/Les-Outils-de-pilotage/Prospective-2050> (accessed 9/01/2023).

10. <https://www.inrae.fr/actualites/pesticides-agriculture-environnement-reduire-lutilisation-pesticides-limiter-impacts-environnementaux> (accessed 9/01/2023).

**Figure 2. Geographic scope of the CSA across the land-sea continuum**



## CSA principles

A CSA's purpose is to establish an inventory and critical analysis of available scientific knowledge at the global level on subjects with multiple dimensions. This analysis is carried out by a committee of scientific experts from public research or higher education institutions. In addition to an overview of the environmental contamination by PPPs and its effects, this assessment also analyses methods, their diversity and areas of applicability, and the development of innovation in this field. By updating the knowledge acquired, the areas of uncertainty and controversy, as well as the questions for which knowledge remains insufficient, this work is intended to inform various stakeholder groups on how to address the impacts of PPPs on biodiversity and ecosystem services from a public policy perspective. It thus contributes to the mission of the research organisations to contribute to public policy. The CSA process is based on INRAE's 'Guidelines for the Conduct of Collective Scientific Assessments and Advanced Studies'<sup>11</sup>. Experts are selected on the basis of their publications in peer-reviewed scientific journals, while ensuring that links of interest (e.g. funding, intellectual affinities, collaborative links), which are inevitable in targeted research, are balanced within the collective, and excluding cases of conflict of interest. Transparency is ensured by describing, within the CSA extended report, the sources and methods used. This CSA was

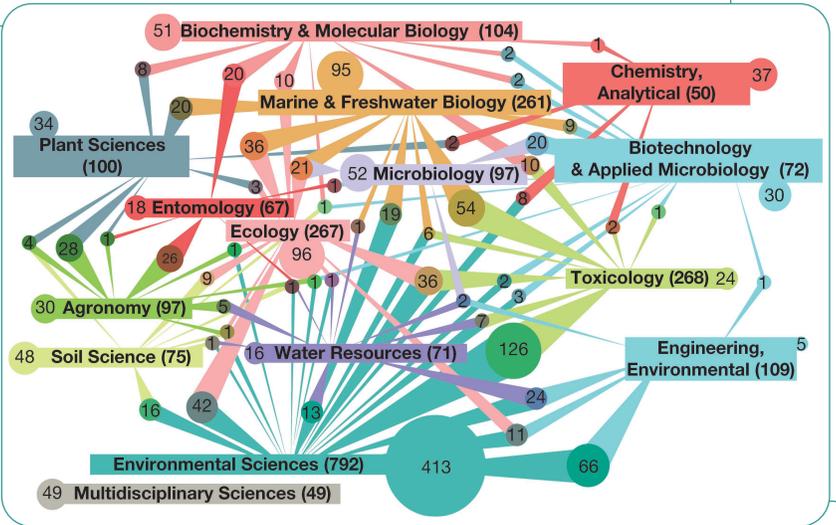
11. [https://www.inrae.fr/sites/default/files/pdf/DEPE\\_Principes\\_Conduite\\_ESCo\\_Etudes\\_V2\\_20211110.pdf](https://www.inrae.fr/sites/default/files/pdf/DEPE_Principes_Conduite_ESCo_Etudes_V2_20211110.pdf) (accessed 9/01/2023).

conducted in collaboration with a stakeholder advisory committee that brings together the main stakeholders involved in the issue of PPP impacts on biodiversity and ecosystem services.

### Composition of the expert group

The expert group was recruited on the basis of an initial search of bibliographic databases to encompass the diversity of topics covered by this CSA. It was headed by three scientific leads: Laure Mamy and Stéphane Pesce, from INRAE, and Wilfried Sanchez, from Ifremer. The 46 researchers (including the leads) involved in the CSA come from 19 research organisations. At the beginning of the CSA process, these 46 experts had authored a total of 1,875 publications indexed in the Web of Science™ (WoS) bibliographic database across a range of research fields (Figure 3). These fields are based on the WoS categories for scientific journals. The majority of experts published in environmental sciences and ecotoxicology fields, with biology of organisms, chemistry and agronomy also represented. Publications in the humanities and social sciences are less commonly referenced in the WoS and are therefore underrepresented in this figure. However, these disciplines are also represented in the CSA by two economists, two legal experts, one sociologist and one anthropologist.

**Figure 3. Clustering of the top 15 Web of Science™ (WoS) categories among the 1,875 expert publications at the onset of the CSA**



In brackets: number of publications classified in the given WoS category.  
 Disks: number of publications ranked within the two linked WoS categories simultaneously.  
 Graph produced with Intellixir©.

## Sources used

This condensed report is based on the findings described in the CSA extended report, which contains the entire bibliography used in the assessment. It is not cited here, except when the data or examples mentioned are taken directly from a publication.

The bibliographic corpus was compiled by searching the WoS and Scopus bibliographic databases, and the Cairn, Springer and Sage platforms for the humanities and social sciences. This initial selection of articles was then completed according to the experts' disciplinary skills.

The bibliographic search focused on the years 2000-2020, in order to update the knowledge acquired since the 2005 CSA 'Pesticides, Agriculture and the Environment'. The geographical scope of the contamination inventory was limited to France and its overseas territories. For the effects of PPPs on biodiversity, ecosystem functions and services, all knowledge from studies conducted in other countries that could be applied to the French context (e.g. types of climate, PPPs, or organisms) were also examined. The bibliographic search was completed, where necessary, with articles from before this period that are fundamental to the understanding of current knowledge, or when the subjects were insufficiently covered by the literature of the last twenty years. It was also updated during the course of the assessment (year 2021 and early 2022), on the basis of the experts' competence and the bibliographic monitoring carried out on the WoS by the CSA librarians. Additional information was provided outside the academic field, including reports produced by institutions using data sources relating to the monitoring of PPP sales or environmental surveillance. With regard to non-agricultural areas, very little academic work deals specifically with these areas and uses. For this section, we mainly used studies that were not published in peer-reviewed scientific journals. These were carried out, depending on the case, under the aegis of the managers of these areas, local decision-makers or other public authorities.

The total number of references cited was 4,460, of which 14% were literature reviews and meta-analyses. Seventy per cent of these references were published in the last ten years. This bibliography covers a wide range of research areas, as shown by the top 15 research fields in which the 3,343 references in the CSA bibliographic corpus that are published in WoS-ranked journals are classified (Figure 4).

**Figure 4. Research fields of the 3,343 references classified in the Web of Science™ (WoS) categories (top 15 categories)**

1,700 Environmental Sciences	357 Entomology	165 Agriculture Multidisciplinary	165 Biodiversity Conservation	152 Engineering Environmental
512 Toxicology	227 Agronomy	147 Multidisciplinary Sciences	116 Plant Sciences	102 Soil Science
540 Ecology	172 Marine Freshwater Biology	126 Biotechnology Applied Microbiology	87 Water Resources	72 Microbiology

## Analysis framework

### I Comprehensive approach to biodiversity

Biodiversity is considered here in the sense of 'biological diversity' as defined by the Convention on Biological Diversity (CBD; United Nations, 1992) as "the variability among living organisms from all sources including, inter alia, terrestrial, marine and other aquatic ecosystems and the ecological complexes of which they are part; this includes diversity within species, between species and of ecosystems". Biodiversity is also considered in terms of population dynamics and flows, interactions, ecological processes and ecosystem functions.

Addressing biodiversity as a whole raises the question of the delimitation of fields of knowledge. Indeed, living environments are made up of biotic (organisms) and abiotic (e.g. minerals, gases) components organised at different scales (e.g. individual, population, ecosystem), which interact with variable temporal dynamics, and fulfilling functions that result from biological activity that enables them to be perpetuated. The keys to analysing such an ensemble can be broken down by environment, type of organism, type of ecosystem, type of interaction, etc., with each typology having its own advantages and limitations, particularly in terms of disciplinary separation.

In addition to this complexity, PPPs can also be characterised by a wide range of attributes: chemical family (e.g. organochlorines), mode of action (e.g. photosynthesis inhibitors), target organisms (e.g. insecticides), use (e.g. fruit production, cereals), toxicological classification (carcinogenic, mutagenic, toxic to reproduction, or CMR level 1 or 2), regulatory category (e.g. basic substances, of concern, low risk, candidate for substitution), and the regulatory status (approved or not approved), etc.

In order to address the issue of impacts on biodiversity as closely as possible to situations as they occur in reality, the full range of impacts of PPP applications and their consequences was considered in the analysis. The substances were therefore not specifically targeted *a priori* in the literature search. However, in order to answer questions relating to certain substances or themes that have been the subject of specific political initiatives over the last decade (chlordecone, copper, glyphosate, neonicotinoids, endocrine disruptors, pollination, SDHI), the CSA report contains appendices that bring together all of the information on these subjects, based on the analyses carried out by the experts.

### I Reference framework for ecosystem functions and services

A common framework has been developed to group the ecological processes potentially impacted by PPPs into 12 categories of ecosystem functions (see section 'Consequences for ecosystem functions'), with the initial aim of linking them to the ecosystem services they support. The reference framework used for ecosystem services is the latest version of the CICES (Common International Classification of Ecosystem Services)<sup>12</sup>. This conceptual framework allowed a common vocabulary to be established at the CSA level, facilitating the synthesis of results. It also made it possible to note the difficulty of linking all

12. <https://cices.eu/>

of the identified ecotoxicological processes with the evaluation of ecosystem services in a comprehensive manner, especially since these two aspects come under different scientific disciplines. The dynamics of the response of ecosystems to the pressures exerted by PPPs, which vary according to time and spatial scales, are therefore difficult to consolidate in the form of impacts measured as a whole on all ecosystem services.

## **I Analysis focusing on studies under realistic environmental conditions**

The existence of a regulatory framework for the placing of PPPs on the market leads to the production of scientific knowledge on their ecotoxicity, thus documenting the assessment of the risks that their use may pose to the environment. This abundant body of knowledge is essentially based on standardised experimental approaches, supplemented by the use of numerical models, and forms the basis for regulatory decisions. The scope and limitations of such an assessment framework are themselves the subject of scientific publications that study the inadequacies of these approaches for estimating impacts at the scales of biodiversity and ecosystem services.

To compile the corpus analysed in this CSA, priority was given to studies that were as integrative as possible and as realistic as possible from an ecological perspective. For example, results from single-species tests have not been systematically reviewed, and are only used insofar as they provide explanations for phenomena observed or suspected under realistic environmental conditions.

## **I Thematic breakdown and cross-cutting themes**

The thematic breakdown presented in Figure 5 was based primarily on the experts' knowledge, and facilitates the compilation and analysis of the bibliographic corpus.

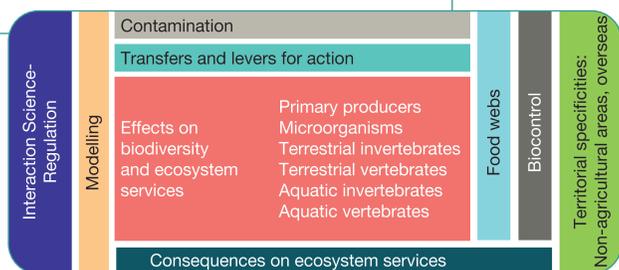
Within the ecotoxicology field, knowledge was analysed by the type of organism (primary producers, i.e. photosynthetic macro- and microorganisms; non-photosynthetic microorganisms; invertebrates; vertebrates), and by the type of environment (terrestrial or aquatic) when this is highly specific (for invertebrates and vertebrates). Focus was also given to the dynamics within food webs, which cross these divisions by type of organism and habitat and play a significant role in both the transfer of substances and the propagation of their effects.

The corpora dealing with contamination, the dynamics of transfer, and the physicochemical transformation of substances, as well as modelling tools, were analysed in a transversal manner. The specificities of biocontrol required a two-pronged approach within this field: in the case of natural substances, they are treated in the same way as other substances, but in a more specific way in the case of living organisms or in studies adopting biocontrol as a separate subject of study (e.g. comparative studies).

As regards non-agricultural areas, knowledge has, as described above, mainly been gathered from non-academic sources, and supplemented by the few scientific studies related to this type of area.

The specificities relating to overseas territories were explored within each of the previously compiled thematic corpora.

**Figure 5. Thematic breakdown of the CSA**



Ecosystem services are the subject of a specific body of literature, and this was analysed as such. Conceptual framing was conducted in order to establish a relationship between the results from the corpus on ecosystem services and those from the analysis of effects on ecosystem functions (examined in the field of ecotoxicology).

Finally, the field of knowledge addressed in this CSA has the distinctive feature of being partly produced within frameworks standardised by regulations (e.g. studies based on data from monitoring imposed by regulations on the surveillance and protection of biodiversity), or for decision-making purposes within regulatory frameworks (e.g. scientific opinions from EFSA or Anses). These interactions between scientific processes and regulatory approval processes partly underlie the scientific dynamics observed in the corpus of this CSA. A multidisciplinary group, involving researchers in law, sociology, ethnology and ecotoxicology, was dedicated to synthesising the scientific work that examines these interactions between science and regulation, particularly in the field of PPP risk assessment.

### Treatment of agricultural practices

This CSA does not address existing tools for limiting the use of PPPs. Topics such as strategies to protect crop health without resorting to PPPs, or the comparison of the impacts on biodiversity of different types of agricultural systems that do or do not use PPPs, are not the subject of this assessment, in order to avoid redundancy with parallel studies. Such complementary analyses have been conducted within the CSA focusing on the natural regulation of pests and diseases, as well as within the *Growing and Protecting Crops Differently* priority research programme. The expert group and corpus topics were not designed to cover these topics, particularly in the field of agronomy, which is not a key element in the approach taken. However, certain methods of using products influence the dispersion dynamics of substances and the exposure of non-target organisms. The available knowledge of how parameters such as application equipment, practices that determine soil conditions, and the adjustments that can be made at the plot and landscape levels influence the impact of PPPs was therefore included in the scope of the study.

# 1. Preamble regarding the fragmentation of knowledge

Despite the size of the scientific corpus dealing with the impacts of PPPs on biodiversity, ecosystem functions and ecosystem services, an examination of the available knowledge quickly reveals the difficulty of generalising results from knowledge that is particularly discontinuous and heterogeneous. This fragmentation of knowledge is partly linked to the topics studied, whether PPPs or biodiversity, which cover a wide range of entities (e.g. substances, transformation products, species, habitats), many of which are not known or not covered in the scientific literature. It is also linked to the diversity of environmental conditions (e.g. pedoclimatic, hydrological) and practices (agricultural or environmental management), which makes generalisation even more difficult. Finally, the frameworks around the production of science have their own constraints and limits, which are not specific to the corpus analysed here, but which must be considered in the critical analysis of the results.

## Patchy and heterogeneous nature

### With regard to substances

Approved or previously approved substances are well known because of the regulatory framework. In 2022, approximately 450 substances were approved at the European Union (EU) level<sup>13</sup>, of which less than 300 were valid for French territory. These substances are used in the composition of more than 1,500 commercial products whose marketing authorization is granted at national level, and whose sales are subject to mandatory reporting. However, fundamental knowledge of the pressure on ecosystems is still lacking, such as a geographic history of applications, possible fraudulent uses, the extent of transfers of substances and their transformation products in the environment, whether within the same environment, between environments (e.g. from soil to surface water or groundwater, from inland waters to the marine environment), or within or between organisms (e.g. within food webs). There are major differences in data availability between environments and between matrices, particularly depending on whether or not they are subject to regulations that require monitoring to be conducted. Aquatic environments are subject to monitoring of the chemical and ecological quality of water bodies, as required

13. [https://ec.europa.eu/food/plants/pesticides/eu-pesticides-database\\_en](https://ec.europa.eu/food/plants/pesticides/eu-pesticides-database_en) (accessed 9/01/2023).

by the European Water Framework Directive (WFD; 2000/60/EC) and the Marine Strategy Framework Directive (MSFD; 2008/56/EC). Despite recent improvements, there are no equivalents for terrestrial and atmospheric environments.

These elements are therefore addressed in the corpus on a case-by-case basis, according to specific research objectives. Thus, the extent of current knowledge remains very uneven depending on the substances considered and the hydro-morphological and geographical contexts in which they are studied (see section "Depending on the context").

The historical perspective is a primary factor in the development of knowledge. This explains the fact that older PPPs, many of which are no longer approved for use today, are better documented than the most recently developed products, for example in the area of biocontrol. Thus, the effects of PPPs are unevenly documented depending on the type of substance, as follows in descending order: organic compounds that are relatively hydrophobic and/or older, inorganic compounds, organic compounds that are less hydrophobic and/or more recently developed, macroorganisms, microorganisms, natural substances, and finally semiochemicals.

A significant knowledge discrepancy can also be seen between the number of substances likely to be found in the environment (those currently on the market and those that were marketed in the past and are persistent, either as such or via their transformation products), those that are looked for, those whose presence is actually detected and those whose effects have been studied.

The spectrum of substances investigated in the environment also varies greatly depending on the matrix concerned. Knowledge of contamination is most abundant in inland waters, followed by marine waters (coastal waters being more closely monitored than offshore waters); there is less knowledge of contamination of the atmosphere and soil. There is also a high degree of variability in knowledge about the contamination of living organisms (biota), with a few taxa, generally used as indicator organisms, being very well studied, while the majority are poorly studied, in a very patchy manner, or not studied at all.

## ■ Regarding biodiversity

Biodiversity is a concept that covers a multitude of study areas: genes, species, ecosystems and interactions, many of which are still little known or unknown. Although it is always tricky to assess knowledge gaps in biodiversity, the proportion of described species in relation to the total number of existing species is estimated to be around 20% at best<sup>14</sup>. According to the Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services (IPBES, 2019), 86% of the world's species and 91% of species in the oceans have not yet been described.

There is no simple indicator for assessing the state of biodiversity. The European WFD and MSFD directives establish monitoring of the ecological status of water bodies, but,

14. <https://theconversation.com/biodiversite-combien-de-millions-despeces-61875> (accessed 9/01/2023).

as with the various substances, the knowledge gathered is very uneven depending on the environment, and only addresses biodiversity in a very fragmented manner.

The geographical areas closest to the sources of contamination are also the most studied in the framework covered by the CSA. Studies are therefore more comprehensive in agricultural areas than in non-agricultural areas, and in coastal marine areas than in offshore or deep-sea areas. Certain types of agricultural settings are more studied, such as field crops, viticulture and arboriculture, while market gardening is less represented, and forestry and grassland even less. This gradient also corresponds in part to the degree of intensity of PPP use in the different crop types.

The effects of PPPs are unevenly documented by organism type, with some groups or species being studied more often, such as honey bees.

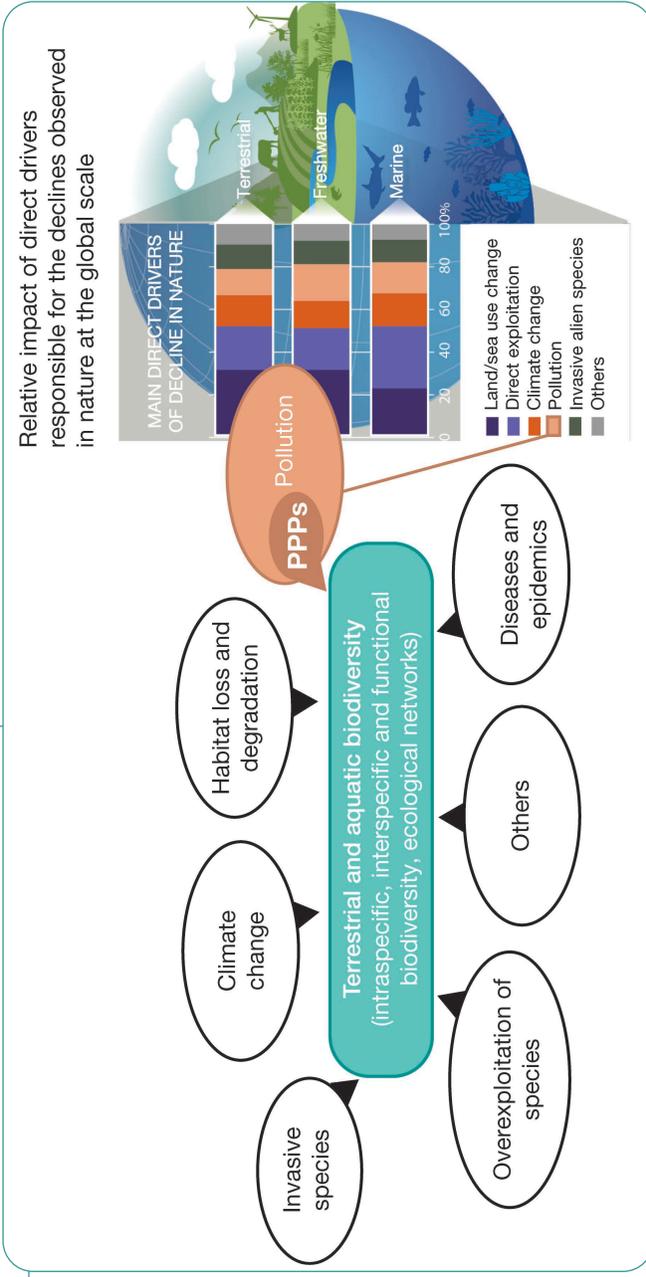
## **I Depending on the context**

The question of the impacts of PPPs on biodiversity assumes that the application of PPPs is known and characterised, as well as the components of biodiversity that are exposed to these products, but also that the initial state and resilience capacity of this biodiversity are known. However, the pressure exerted by PPPs is a dynamic phenomenon that depends on the location and repetition of treatments, with the same or a variety of PPPs, the transfer of substances into the environment and their transformation. It is thus characterised by a spatial and temporal dynamic specific to each context. Similarly, biodiversity is a constantly changing entity under the effect of a multitude of biotic and abiotic factors and pressures (Figure 6), the individual effects of which are difficult to isolate, especially as most of them interact with each other (e.g. pollution by PPPs can contribute to the degradation of habitats; climate change can favour the development of certain invasive species). The impacts of PPPs are one factor in this process, and induce responses that lead to other effects at multiple temporal and spatial scales. At the global scale, IPBES (2019) has broadly ranked the different drivers of nature decline. Pollution, which includes PPPs, appears to be the third (in freshwater) or fourth (in terrestrial and marine environments) cause of the observed declines, depending on the environment. However, this hierarchy may differ depending on the situation viewed at a smaller scale.

Of all the possible exposure scenarios, the scientific demonstration of the impacts of PPPs can only be established for a small number of substances, organisms and types of effects observed, in an equally small range of environmental contexts. With regard to the dynamics of trophic transfers of substances, for example, studies document certain combinations (molecule × resource × consumer organism) which correspond to a tiny fraction of the range of possibilities.

This strong dependence on the setting limits the potential for generalising from the available knowledge. Thus, only by combining scientific results from complementary approaches can an inventory of the field be established.

**Figure 6. Illustration of the different pressures that can affect biodiversity (source: IPBES, 2019)**



## **Complementarity of approaches and objects of study**

Whether a focus is on the degree of contamination of environments, transfers, exposure, the effects of contamination and their consequences on biodiversity or the functioning of ecosystems and the services they provide, no single scientific approach is capable of documenting all factors, species, environments and dynamics at play. The complexity of interactions between PPPs and biodiversity can only be addressed by combining different approaches, adapted according to the context and the objective pursued, but at the risk of contributing to the heterogeneity of the scientific methods used and data generated. Moreover, the selection of topics on which research has been published is the result of choices whose determinants may lie outside as well as within the academic sphere. These must be taken into account when interpreting the results. The CSA does, however, make it possible to identify converging elements.

### **Tools and investigative scales: the field, mesocosms, laboratories and modelling**

Depending on the research objectives, a study may combine different types of approaches, including direct observations in the field, experiments in mesocosms or microcosms based on controlled variation of certain factors, laboratory studies on model organisms and modelling approaches. These approaches are of course interdependent, since the parameterisation and validation of the models are based on experimental and observational data, and the choice of variables measured in experiments and observations also depends on the results obtained elsewhere through other studies or modelling. Each of these knowledge-generating mechanisms has its strengths and limitations. For example, the reduction of factors taken into consideration in laboratory studies improves the robustness of the evidence of established causality, but alters their capacity to represent the reality of field situations.

The existing body of knowledge is thus very heterogeneous, even for the same substance and the same organism, depending, for example, on the type of effect studied, the observation conditions, or the level of biological organisation considered.

Some harmonisation initiatives, which are still relatively rare, have been undertaken to standardise the conditions for highlighting the effects of substances in the natural environment, in order to be able to capitalise on the results of different studies. On the other hand, the diversity of the subjects studied and of the observation conditions is limited. Therefore, a complementarity can be found along the gradient linking standardised systems, which can reproducibly establish the relationship between a characterised exposure and a measured effect criterion, and non-standardised systems, the results of which cannot be generalised but which are likely to highlight correlations and identify previously neglected research questions (e.g. little-studied environments, exposure route, type of effect, type of organism, measurement method).

## I Choice of study subject and publication

Given the wide scope of the area of investigation that biodiversity constitutes, this scientific approach selects study subjects on the basis of various criteria: identified issue (e.g. pollination, biological control, emblematic species), particular sensitivity/tolerance to PPPs (e.g. indicator species, sentinel species), place or role in the ecosystem (e.g. focal species, keystone species, ecosystem engineer), ease of study (e.g. ease of observation, short life cycle, suitability for laboratory experiments), environmental and ethical impacts of the study (e.g. deliberate contamination for *in situ* testing, animal suffering). The factors leading to the production of scientific knowledge on certain topics rather than others can be found outside the scientific field, as well as in the academic sphere itself.

### Factors outside the scientific sphere

The factors underlying the choice of subjects studied may stem from direct interests, which are examined in terms of the links of interest of the researchers or research teams conducting the research. The orientation of the research may be influenced by the 'emblematic' character of certain substances or organisms (e.g. glyphosate, bees and pollination), or by the perception of their usefulness (e.g. pollinators, predators of crop pests). The literature analysed in the humanities and social science field reveals this phenomenon of 'mutual emulation' between different components. For example, a coalition of stakeholders is aware of concerns relating to a type of substance or organism leads to a scientific dynamic that documents these concerns. Consequently, a demand for expertise or research incentives from public authorities that may precede or follow this expertise then occurs, and regulatory decisions are then made based on this scientific knowledge. This results in 'fashion effects' in scientific production, with certain subjects being more intensively investigated at certain times.

### Factors relating to the broader scientific domain

Within the broader academic setting, biases that should also be considered when analysing articles are routinely identified. In particular, the so-called 'publication bias' refers to the fact that results that fail to show an effect (either because there is no effect or because the research design did not enable the effect to be shown) are published less than results that do show an effect. The reluctance of researchers to exploit results that do not report an effect, and the reluctance of journals to publish them, generally leads to a scientific output in ecotoxicology which is biased towards the demonstration of effects.

Similarly, the so-called 'streetlight effect' illustrates the fact that a result can only be obtained in the area illuminated by research already conducted. Research tools, be they concepts, methodological approaches or measuring instruments, inevitably partly predetermine the result that can be obtained. Thus, newly identified effects of PPPs are not necessarily new effects, but effects that the previously implemented research tools were unable to reveal.

Finally, every scientific result has a range of validity and is bounded by margins of error that accompany the measurements and calculations. Although this information tends to be better documented in the published literature, uncertainties generally remain insufficiently described. For example, the protocols for acquiring data on PPP concentrations in the environment and the quality of approaches used (e.g. blank samples, analytical tracers, development and validation of methods) are increasingly detailed (from sampling, through the analytical stage, to the results) and the data are generally available in the appendices of publications. However, data on measurement uncertainties (including or excluding the sampling step) are still very patchy.

## ■ Identification of clusters of convergence

The fragmentary and heterogeneous nature of knowledge in this domain prevents results from being aggregated in the form of indices or common, systematic quantification. On the other hand, the complementarity of approaches and the diversity of study subjects make it possible to identify clusters of convergence between different results. Studies based on analysing the correlation between population trends and PPP use, but without demonstrating causality on a large scale, are compared with other studies carried out under controlled conditions that do identify causal links. This synthesis of results, which draws on the diverse skills and contrasting perspectives of the experts involved in this CSA, enables us to establish the current state of knowledge, including the main disagreements and shortcomings, as revealed by the bibliographic corpus.



## 2. Environmental contamination by PPPs and exposure of organisms

The degree of contamination of the environment by PPPs is difficult to characterise in an overall manner, as this contamination can vary greatly in time (e.g. peaks following the application of treatments or a weather event), in space, and according to the compartment under investigation (soil, air, water, sediment, biota). Moreover, contamination is analysed using methods that are themselves constantly being improved, which does not always permit the identification of major trends. The data generally concern the main substances used in volume, those expected to be present in the investigated compartment according to existing hypotheses regarding their fate, or those identified as the most toxic. The results presented in this section provide a first step towards assessing current trends, bearing in mind that they generally underestimate the presence of PPPs that are not widely sought, including PPP transformation products, and PPPs present in quantities below the detection capacity of existing methods at the time of the study. However, the available knowledge show evidence of contamination of all matrices and environments by a wide variety of substances, particularly in agricultural areas. However, this contamination also spreads to other areas due to transfer dynamics and the transformation of substances after their application. This leads to the exposure of organisms in a variety of environments (terrestrial and aquatic) according to processes that are highly dependent on the context. However, certain measures can limit this dispersion and the resulting exposure routes. Finally, knowledge of the dynamics of contamination and exposure of organisms could be improved through research that has already been undertaken or is planned.

### Proven environmental contamination by a wide range of PPPs

Environmental contamination by PPPs is assessed using constantly evolving sampling strategies and analytical techniques. This development allows, on the one hand, regular increases in the number and diversity of substances analysed in the various environments and matrices and, on the other hand, a reduction in the thresholds for detection and quantification. Analysis of scientific literature and available data shows that all environments, whether terrestrial, aquatic (continental or marine), as well as the atmosphere, including the biota present in these different environments, are contaminated by a wide variety of substances and transformation products (Figure 7). Figure 8 shows a

schematic overall breakdown of this contamination in mainland France and the French overseas territories. This is based on an assessment, from the studied literature, of the contamination gradients between different substances and between different environments. Overall contamination is in fact difficult to quantify more precisely, as it is still largely unknown (see section ‘With regard to substances’) and varies greatly in space and time. This variability is initially described, and then the principal results concerning the substances most frequently found and the most contaminated environments, particularly in agricultural areas, are compiled.

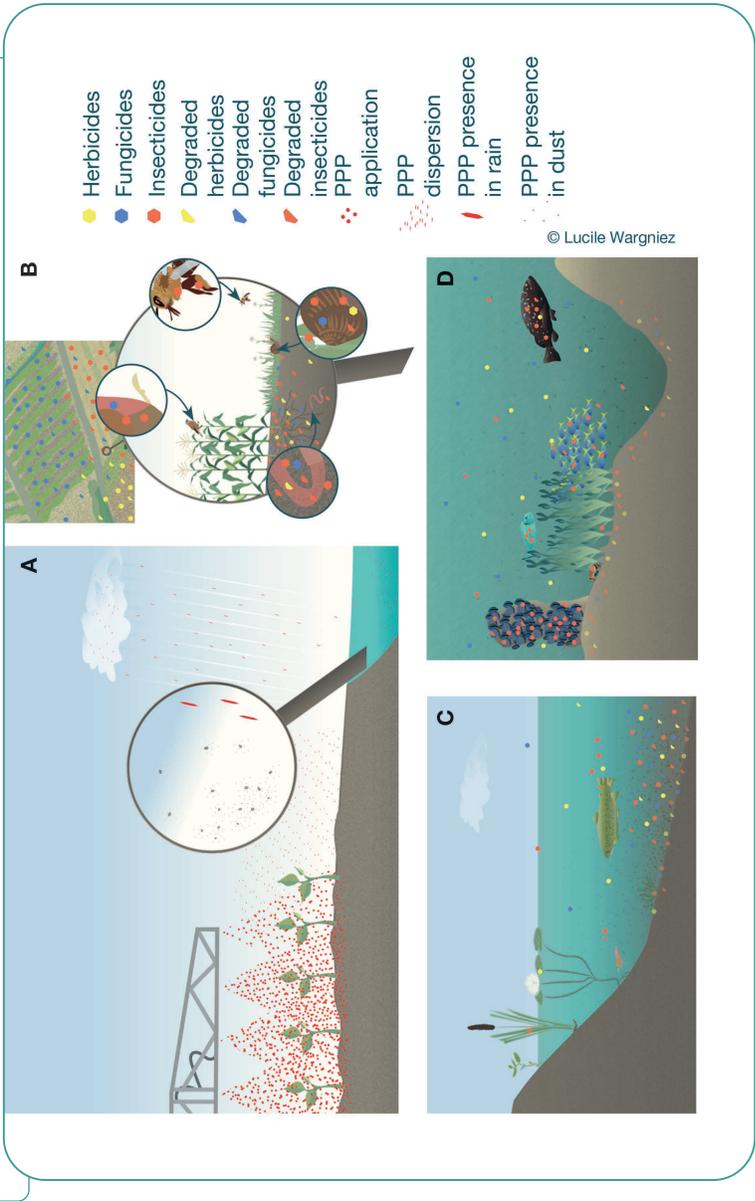
## I Spatial and temporal variability of contamination

The spatial variability of substance classes and measured concentrations is mainly related to the geographical proximity of the application site, the type of compartment considered (e.g. soil, surface water, sediment, air, and biota) and the physico-chemical characteristics of the substances, which predispose them, for instance, to associate with certain elements or to degrade. Persistent organic compounds (most of which are now banned, mainly insecticides, some herbicides and the fungicide hexachlorobenzene, or HCB) are found in most matrices and more specifically in sediments and biota. Hydrophilic compounds (e.g. many herbicides) are mostly found in water. All families of organic PPPs (herbicides, fungicides or insecticides) are found in the atmosphere. With the exception of the studies devoted to the contamination of certain overseas territories by chlordecone (Della Rossa *et al.*, 2017), very few existing references aim to jointly characterise the PPP contamination of different environments and/or different matrices in the same environment (e.g. physical environment vs. biota; sediment vs. surface water; soil vs. aquatic environment along a continuum).

The short-term temporal changes in ecosystem contamination by PPPs depend in particular on the rate and intensity of application, and on soil and climatic conditions that affect the degradation, bioavailability and transfer of these substances. Longer-term changes can result from significant change in land use and management (e.g. establishment of buffer zones to reduce transfer) and PPP use. Some of these changes may result from regulatory measures, such as the banning of certain substances or the limitation of PPP uses in certain areas, notably following the 2017 implementation of the Labbé law (République française, 2014) which concerns non-agricultural uses.

The scientific literature does not generally provide information on temporal changes in contamination beyond two or three years, a time span rarely exceeded in academic studies. Moreover, it generally considers only a limited number of substances (Chow *et al.*, 2020). However, thanks to long-term regulatory monitoring and surveillance networks, it is now evident that the concentration levels of banned substances decrease in aquatic environments and in the atmosphere after their use has been stopped. This leads to an overall decrease in PPP concentrations in aquatic environments, based on the most frequently sought substances.

Figure 7. Distribution of PPP contamination in different matrices (including biota) in the atmosphere (A), terrestrial (B), inland waters (C) and marine environments (D)



**Figure 8. Environmental contamination by PPPs**  
(adapted from Pesce *et al.*, 2023a)



## Substances most frequently found

### In soils

The presence of PPPs is reported in the vast majority of soils, particularly in agricultural soils under conventional farming practices, which are the most studied.

Thus, most studies aimed at quantifying a wide range of PPPs in these agricultural soils report the presence of a wide variety of substances, mostly present in mixtures. For example, in soils sampled in the French Deux-Sèvres region in winter cereal plots, Pelosi *et al.* (2021) detected an average of 11 PPPs (i.e. 35% of the PPPs tested), with 83% of the soils containing at least 5 PPPs. In this study, soils from organic farming plots contained an average of 6 PPPs (potentially as a result of aerial or waterborne transfer), those from grasslands between 1 and 16, and those collected under hedgerows between 1 and 17. In another recent Europe-wide study (11 countries), Silva *et al.* (2019) detected mixtures of 2 to 10 PPPs (based on 76 substances tested) in 55% of the 30 soils sampled in France, with total concentrations above 0.5 mg/kg and 1 mg/kg in 7% and 3% of cases, respectively. However, the quantities and diversity of PPPs in soils vary greatly between studies, and in particular between crops. The number of studies (and detections) is higher for certain well-known compounds, such as the neonicotinoid insecticide imidacloprid, the insecticide chlordecone in the French Caribbean or the fungicide copper. In a large study of 74 soils in mainland France, Bonmatin *et al.* (2005) detected imidacloprid at a concentration higher than 0.1 µg/kg in 91% of the soils (excluding organically grown soils, in which no

traces were detectable). Chlordecone, widely used until 1993 to control the banana weevil, is also omnipresent in the soils of Martinique and Guadeloupe located in or near banana plantations, with contamination levels sometimes exceeding 1 mg/kg (Desprats, 2020). Similarly, other PPPs that were banned many years ago are still present in French soils (e.g. dichlorodiphenyltrichloroethane, or DDT, lindane, atrazine). Copper is also the subject of numerous studies, and its concentration levels in soils have been mapped as part of the Soil Quality Monitoring Network for French soils *Réseau de mesure de la qualité des sols* (RMQS). Its concentration commonly reaches several hundred mg/kg in vineyard soils, where this substance is omnipresent.

The analysis of the literature did not provide information on the status of contamination of the various environments by biocontrol products. This issue is virtually ignored in the international scientific literature except for very few studies, mostly conducted outside of France (Espinasse *et al.*, 2003), concerning the fate of *Bacillus thuringiensis* (Bt) proteins in the soil. These studies indicate that Bt toxins could remain biologically active even after adsorption to the soil, especially to clays, where they are strongly bound and less rapidly degraded than their free form (Liu *et al.*, 2021). However, the origin of the toxins is not distinguished between PPP and vector control uses. The same applies to biocontrol substances that may be naturally present in the environment (e.g. fatty acids, potassium hydrogen carbonate, aluminium silicate, sulphur). However, some publications have shown that certain substances may persist in the environment (abamectin, paraffin oil, spinosad, phosphonates).

### In inland and marine environments

The substances identified as priorities by the WFD are more frequently sought. They are regularly detected in the various matrices making up these environments, and this observation concerns several substances which have been banned in France for many years, confirming their persistence in the environment. This is the case, for example, of certain triazine and phenylurea herbicides (e.g. atrazine, simazine, terbutryn, diuron, isoproturon).

The herbicide glyphosate and its transformation product, aminomethylphosphonic acid (AMPA), are not considered to be priority substances in the regulatory sense, but they are nonetheless the subject of a large number of studies in inland aquatic environments, in which they are very frequently detected. The meta-analysis by Carles *et al.* (2019) based on more than 72,000 data points from monitoring programmes conducted between 2013 and 2017 showed that glyphosate (limit of quantification, LOQ = 0.03 µg/l) and AMPA (LOQ = 0.02 µg/l) were quantified in 43% and 63% of surface water samples respectively, with average concentrations in France between the LOQ and < 0.4 µg/l for glyphosate and between 0.2 µg/l and > 1 µg/l for AMPA. Carles *et al.* (2019) showed that in French surface waters AMPA is more frequently quantified than glyphosate. Conversely, in the marine environment, glyphosate and AMPA remain relatively undetected to date and are only detected very occasionally, generally in transition zones (e.g. estuaries, downstream areas of rivers), but at sometimes high concentrations (which can exceed 1 µg/l) compared to other herbicides detected in these environments. However, it is important

to note that AMPA is also a transformation product of aminomethylene phosphonates, notably used in detergents. Its presence in aquatic environments is therefore not exclusively linked to the use of glyphosate.

Copper is the subject of particular attention in inland and marine aquatic environments, where recorded concentrations regularly indicate contamination by this metal in surface waters, sediments and biota. In inland aquatic environments, copper concentrations are in the order of a few  $\mu\text{g/l}$  to a few tens of  $\mu\text{g/l}$  in surface water, and in the order of several tens to several hundreds of  $\text{mg/kg}$  in sediments. Soil erosion, particularly in vineyards, appears to be a major source of copper in these environments, although it may also have a natural origin or be linked to uses as a biocide. In the marine environment, copper is also detected in all matrices (i.e. in surface water, sediment and biota), but its use as a biocide in antifouling paints is a confounding factor that makes it difficult to determine the origin of this pollution. The use of isotope analysis offers good prospects in this area. In the French Caribbean, studies on the presence of chlordecone in aquatic environments reveal persistent contamination of all matrices (surface water, sediments and biota) in rivers and the marine environment, with a decreasing gradient from the coast to the open sea, as well as bioaccumulation in marine fauna.

With the exception of tebuconazole (and other fungicides of the triazole family) and copper, herbicides (including certain active substances that have been banned for many years) and their transformation products are the substances most often detected and quantified in high concentrations in the surface waters of continental and marine aquatic environments, with concentrations often ranging from a few  $\text{ng/l}$  to a few hundred  $\text{ng/l}$  (certain contamination peaks can reach a few  $\mu\text{g/l}$ ). However, concentrations are lower in the marine environment. Thus, 75% of the levels of dissolved PPPs measured in coastal waters are less than  $50 \text{ ng/l}$ , but as we move closer to the coasts and therefore to the sources of these substances, both the number of substances measured simultaneously and their concentrations increase.

In contrast, organochlorine PPPs, which belong to the POP (persistent organic pollutants) group and are now banned, are very rarely found in water. However, their physico-chemical properties and persistence mean that they are predominantly concentrated in sediments and biota, in which they are sometimes quantified at several  $\text{mg/kg}$ , or even several dozen  $\text{mg/kg}$ . These substances are therefore specifically monitored to assess the contamination of sediments and aquatic biota, mainly in the marine environment, as these two matrices are seldom studied in inland aquatic environments. As a result, approximately 90% of the organic PPPs detected in marine biota in mainland France are organochlorine POPs (mainly insecticides and the fungicide HCB). This type of contamination also concerns deep-sea organisms, reflecting the ubiquity of PPP contamination, as highlighted by Munsch *et al.* (2019), who reported contamination in deep-sea pelagic organisms by the insecticides DDT, hexachlorocyclohexane (or HCH), chlorinated cyclo-dienes and the fungicide HCB.

## In the atmosphere

The presence of organic PPPs in the atmosphere (measured with airborne gas and aerosol samplers or bioindicators such as pine needles or lichens) is confirmed in both rural and urban areas, regardless of the category of use (herbicides, fungicides or insecticides), the distribution of these categories of use being dependent on the surrounding agricultural context. However, concentration levels vary according to the compounds, the quantities used and the distance to the source. These range from a few  $\text{pg}/\text{m}^3$  to several  $\text{ng}/\text{m}^3$ , or even  $\mu\text{g}/\text{m}^3$  in specific cases, mainly for folpet and more rarely for chlorothalonil, in a limited number of sites located close to treated plots. The diversity of substances as well as the range of concentrations is illustrated in particular by the National Exploratory Pesticide Campaign - *Campagne nationale exploratoire des pesticides* (CNEP, 2018-2019), which involved the monitoring of 75 substances. Its results revealed that 56 substances were quantified in mainland France and 19 in the overseas territories, reflecting the lower diversity of substances used in the latter (Anses, 2020). In terms of maximum concentration, 20 substances were detected with values between 1 and 10  $\text{ng}/\text{m}^3$  and 5 with values between 10 and 100  $\text{ng}/\text{m}^3$ . Several high concentrations, in excess of 100  $\text{ng}/\text{m}^3$ , were observed locally (folpet, pyrimethanil and prosulfocarb). In terms of median values, only 5 substances (glyphosate, lindane, S-metolachlor, pendimethalin and triallate) in mainland France and 2 substances in the overseas territories (S-metolachlor and lindane) showed a non-zero value, indicating that, for all other substances, more than half of the results were below the detection limit. It should be noted that 8 POPs were tested for during the CNEP, including lindane, which has a quantification frequency of over 70%. To date, no traces of chlordecone have been reported in the air in the French Caribbean. With regard to biocontrol, following this campaign, Anses identified air-borne abamectin as a priority for research (Anses, 2020).

## In different environments

In addition to the findings specific to each of the environments described above, it appears that on the basis of the PPPs sought in the environment, the organic PPPs currently detected most frequently (both in terms of frequency of detection and maximum concentrations) among those authorised for use are mainly the triazole fungicide tebuconazole (in all environments), the organochlorine herbicide S-metolachlor (mainly in aquatic environments and the atmosphere) and the organofluorine herbicide diflufenicanil.

## ■ The most heavily contaminated areas

There is a large imbalance between the number of studies involving agricultural areas and those involving non-agricultural areas. Although it is not always possible to quantify the relative share of sources of contamination (especially when moving away from them), agriculture remains the main activity using PPPs, and agricultural areas are the most contaminated by these substances. The use of PPPs in non-agricultural areas may cause local issues (but most of the studies on this subject were conducted before the implementation

of the Labbé law), as well as other uses of the same substances, particularly as biocides (e.g. vector control, antifouling treatments, control of invasive species), which are outside the scope of this CSA. Finally, the discharge of treated effluent (wastewater) from domestic or industrial sources can also be a significant source of pollution of aquatic environments. PPPs sold in France are mostly intended for crop protection. The National Database of Sales by Authorised Distributors *La Banque nationale des ventes par les distributeurs agréés* (BNVD) indicates that the share of quantities sold of active substances intended for the maintenance of non-agricultural areas, including biocontrol products, has fallen from 6% in 2009 to 2% in 2020 of total sales. This proportion is slightly underestimated because products with mixed agricultural and non-agricultural uses are counted as agricultural uses. Notwithstanding, the use of PPPs has been severely restricted by the Labbé law adopted in 2014 and subsequent provisions (see section ‘Requirements and complexity of PPP regulations’). Agricultural land remains the main area of PPP application, with an area of arable land (agricultural area without grassland) of 20 Mha, while non-agricultural areas cover 3 to 4 Mha overall<sup>15</sup>.

For organic PPPs, it is generally difficult to establish specific links between a type of crop and a type or family of substances. To our knowledge, no method is currently available to distinguish the source and type of use (plant protection or biocide) in matrices that are not directly treated with these substances. In the context of air quality monitoring, PPP contamination is observed in both rural and urban areas, sometimes with a higher number of compounds in the latter, but at generally lower concentrations than in rural areas. The influence of agricultural uses on atmospheric contamination is highlighted by the fairly pronounced seasonality of contamination levels for currently authorised PPPs, which depends on the periods and types of treatment, whereas the level of contamination by POPs, which have been banned for many years, is relatively constant throughout the year. The link between the application of PPPs and the air contamination by some of these substances is also supported by the fact that their concentrations in this matrix tend to decrease when moving away from the treatment areas (Coscollà and Yusà, 2016).

With regards to copper, the analysis of the literature clearly shows that viticulture is one of the main sources of contamination due to its use as a PPP. However, viticulture and other agricultural practices involving the use of copper (e.g. olive and fruit cultivation) are not the only sources of environmental contamination by this metal trace element, and other possible sources must also be considered (e.g. biocides, pig manure-based amendments), as well as natural endogenous inputs. Isotope methods have shown their potential to distinguish the origin of copper contamination in the marine environment.

## I Contamination of biota and exposure of organisms

Despite the fact that knowledge is rather fragmentary (in particular concerning currently approved substances, which are not well studied), the literature reports widespread

15. Source: Écophyto monitoring note 2018-2019: <https://agriculture.gouv.fr/telecharger/106541?token=1f-20b7a16e99b1eff3309f39fe68e55147a9ac11b6bb9b46f64b38aa2a6ee652> (accessed 9/01/2023).

contamination of biota, from microbial assemblages to large predators, by a wide variety of PPPs. This confirms the exposure of organisms in different environments.

Except for DDT and other organochlorine PPPs, little work has been done on contamination of terrestrial invertebrates. The use of these organisms in PPP contamination monitoring is therefore limited at present to a few studies concerning, for example, earthworms, snails or bees, which demonstrate that these organisms can be contaminated by currently used PPPs. On the other hand, there is considerable evidence of trophic contamination in terrestrial vertebrates, via the consumption of treated food items (e.g. omnivores and herbivores/granivores, including small mammals and birds) or the consumption of PPP-contaminated prey (e.g. predators and scavengers among wildlife). In aquatic environments, most of the knowledge concerning the contamination of biota by PPPs comes from marine studies. However, available data mainly involves substances that are now banned. For example, various bivalve molluscs (e.g. mussels, oysters) have been widely used for nearly fifty years to assess changes in the contamination of various coastal marine ecosystems by organochlorine insecticides, revealing a significant decrease in their concentrations over the decades, even though these substances are still found. In addition, chlordecone was analysed in more than a hundred different marine species covering all trophic levels in mangrove, seagrass and coral reef ecosystems, from primary producers to marine mammals. This has revealed the existence of a transfer of this substance in marine food webs by bioaccumulation and, sometimes, bioamplification (Dromard *et al.*, 2022). Trophic transfer is mainly described for substances that are now banned, including POPs (e.g. lindane, heptachlor, endosulfan) and their transformation products, as well as some more recently used PPPs (fipronil and diuron) in different environments. For the PPPs currently in use, laboratory studies have revealed, for a very limited number of substances, certain cases of bioaccumulation in organisms, and/or their food items and/or of biomagnification in food webs. In addition to a lack of research on this subject, the small number of studies reporting these phenomena of accumulation and/or biomagnification of the most recent substances can be explained by the fact that the substances currently authorised are less hydrophobic than the older substances now banned.

## Transfer dynamics and fate of substances

During the treatment of crops with PPPs, a portion of the applied amounts does not reach its target and is dispersed in the environment. The mass balance, i.e. the distribution of these quantities between vegetation, air, soil and water, remains uncertain and difficult to establish due to its dynamic nature. Similarly, although measurement and modelling tools have advanced, predicting the fate of a substance in the environment remains very difficult given the multifactorial mechanisms that are involved, which are sometimes antagonistic and heterogeneous depending on the context. This dynamic highlights the key role of connectivity between environments. Although, in general, contamination tends

to decrease with distance from the place of application, certain areas have the capacity to retain or concentrate the substances.

## ■ Multi-factorial mechanisms

Among the main transfer pathways of PPPs in the environment, a distinction is generally made between horizontal transfers (drift, runoff, drainage) and vertical transfers (leaching and volatilization). Transfers can also take place, to a lesser extent, via plants (by root uptake or contact) or animals (by bioaccumulation and biomagnification along trophic chains). The main transfer routes differ significantly between the time of application, with a preponderant share of drift, and the periods that follow, during which waterborne transfer processes (runoff, leaching, and drainage) can become dominant when rainfall or irrigation episodes occur. Their relative importance depends strongly on the agro-pedoclimatic conditions.

Post-application, the proportion of PPP transferred from the soil to the various environmental compartments in relation to the quantity applied is still relatively unknown. However, a few order of magnitude estimates are available: exports outside the treated plot are around 1%, even though, in extreme situations (heavy rainfall just after treatment on low-permeability soil), they can be as high as 15% through runoff, and even as high as 60% through volatilisation.

However, this distribution is highly variable because it depends on many physical, physicochemical and biological factors, as well as agricultural practices, which play a role in both the transfers and their limitations. The most important of these factors are:

- the physicochemical properties of the active ingredients, co-formulants and adjuvants in the products or when used in combination;
- the application strategy and equipment used during application, in relation to the product formulation: spraying methods, burial, etc;
- the crop treated and its three-dimensional structure;
- the weather conditions during and after application;
- the type and condition of the soil: structure, organic matter content, water content, plant cover, microbial activity, etc;
- the pedoclimatic characteristics of the treated area and its landscape layout.

## ■ Interconnected environments

Transfers of PPPs within and between the different physical components of the environment contribute to the spatio-temporal lag between the application of the substances and the exposure of organisms.

At the time of treatment, the drift of PPPs can reach several dozen or even hundreds of metres, as observed for glyphosate (Bernasconi *et al.*, 2021). The transport of molecules from the treated plot and their dispersion in the atmosphere can thus lead to the contamination of neighbouring agricultural plots and untreated environments. In addition

to dry deposition of PPPs in the air in gaseous or particulate form, atmospheric transfers of these substances can occur by wet deposition, in the case of contaminated rainfall. Atmospheric transfers can sometimes occur on a large geographical scale (regional or even continental) when PPPs are highly persistent in this environment.

The transfer of PPPs, including the parent molecules and their possible transformation products, takes place along geographical continuums, from terrestrial ecosystems to inland and then marine aquatic ecosystems, through different mechanisms (e.g. runoff, erosion, entrainment by currents). It has been shown that intensive cultivation in catchment areas contributes to the contamination of the coastal marine environment, with the dilution process tending to reduce the concentrations from upstream to downstream.

PPP transfers through biota may also concern parent molecules or their transformation products. Residue measurements carried out in organisms show an accumulation of a large number of compounds, mainly in the form of mixtures. This accumulation can be a source of trophic transfer of PPPs, which has been notably evidenced in terrestrial and aquatic vertebrates with bioaccumulation or even biomagnification phenomena, particularly for molecules that are now banned but persistent. However, these phenomena do not only concern these banned molecules, as illustrated by the example of pyrethroids (Rasmussen *et al.*, 2013; Pristed *et al.*, 2016).

Some studies have shown that trophic transfers across ecosystems can take place, for example from the aquatic to the terrestrial environment, due in particular to the accumulation of PPPs in organisms during their aquatic life stage, followed by their consumption by terrestrial predators after they have left the water (the case for numerous insect species with aquatic larvae and aerial adults). However, this phenomenon, which has been demonstrated for older POPs and PPPs, remains poorly described.

In general, little is currently known about how organisms are exposed to the new hydrophilic PPPs, particularly with regard to the fate of the substances as they pass through the organism.

Accumulation along aquatic trophic chains also leads to the transfer of coastal marine contamination to predators that develop in environments that are not very contaminated, but which are exposed through the consumption of contaminated prey.

## Influence of context on exposure dynamics

The exposure of organisms is the first step in the processes likely to generate biological effects, and it strongly conditions the dynamics of these effects. It is therefore a fundamental parameter, but one that is difficult to define under real environmental conditions, as it involves the history of contact between an organism (or a group of organisms), with its own dynamics (e.g. life stages, movements, exposure to other stresses), and a substance or group of substances, also with dynamic behaviours (linked in particular to the

various transfer and transformation processes). The exposure of organisms results from their contact with a substance or group of substances in a bioavailable form, i.e. one that can be absorbed and reach a biological target in an active form.

In real-life conditions, exposure is highly context dependent and can vary from one site to another, from one time to another, from one organism to another. The main factors identified as contributing to this variability are related to the dynamics of the substances and the environment, and the degree to which organisms and substances are present simultaneously.

### **Factors relating to exposure and bioavailability of PPPs**

The properties conferred by the physico-chemical characteristics of each molecule (e.g. octanol/water partition coefficient, or  $K_{ow}$ , octanol/air partition coefficient, or  $K_{oa}$ ) determine the persistence, mobility and environmental availability of PPPs in the environment and will therefore influence the exposure of organisms to these molecules. In addition to the characteristics of the active ingredient, the overall formulation in the product used (active ingredient alone vs. liquid formulation and coating or seed treatment) and any associated adjuvants also influence persistence, mobility and availability. Comparing the fate and/or effect of active ingredients in different formulations can thus lead to apparently contradictory conclusions.

These properties are combined with those of the environmental matrices encountered during transfers, in which the parameters commonly identified as most critical to the bioavailability of substances are the amount of water (for soils and sediments), organic matter and clays. Other factors such as temperature, pH or exposure to ultraviolet radiation will also influence the bioavailability and/or transformation of the PPPs in question through abiotic degradation processes, but also through adsorption/desorption phenomena.

For example, it has been shown that copper toxicity in aquatic environments varies according to the amount of dissolved organic matter present, as well as the complexing capacity of the water or the pH. In this environment, temperature can also affect the exposure of organisms to copper, as illustrated by Lambert *et al.* (2016) who demonstrated a reduction in its accumulation in periphytic microbial biofilms following an increase in water temperature.

More generally, the adsorption of PPPs onto organic particles, a phenomenon that depends on the intrinsic properties of each molecule and the characteristics of the matrix (e.g. soil, aquatic sediment, suspended particles), will increase their persistence. This effect is evident, for example, in the atmosphere, where PPPs adsorbed on aerosols can persist for longer periods. In the case of gradual release, this may result in chronic exposure of organisms.

Most of the organic PPPs currently in use tend to be less persistent than the older, now banned chemicals. However, some currently approved substances can persist for several months in soil to which they are applied. For example, it has been shown under experimental conditions that there is a high risk of soil contamination by picloram and other PPPs of the sulfonylurea family more than four months after treatment (Passos *et al.*, 2018). Moreover, repeated applications may result in accumulation and *de facto* persistence (generally referred to as

pseudo-persistence) in the soil. However, in the case of certain PPPs under certain conditions, repeated treatments can promote the development of microbial degradation capacities, due to the regular exposure of soil microorganisms, and thus stimulate the development of biodegradation phenomena that help to reduce the persistence of these substances.

The transformation of PPPs is also an important parameter to be considered with regard to exposure dynamics. It reduces the concentration of the parent substance, but adds the products of its transformation and thus increases the diversity of substances in the chemical exposome. For example, the main transformation product of DDT, dichlorodiphenyldichloroethylene (DDE), which is highly persistent, is now being detected more frequently and in higher concentrations than its parent compound.

### ■ Co-occurrence of organisms and substances

Exposure involves the simultaneous presence of a substance and an organism. It is determined by the conjunction between the temporality of the treatments and transfer processes, and that of the ecology of the species, including their generation rates and life cycles.

Thus, the diversity of ecological characteristics (habitat, phenology) and biological traits, such as feeding, breathing or reproductive patterns, may lead to very different levels of exposure to PPPs between species within the same ecosystem. For example, longer-lived species will tend to have more discontinuous and repeated exposure, including during breeding periods, while shorter-lived species will tend to be exposed more continuously throughout their life cycle, including for several generations (e.g. microorganisms and zooplankton).

Furthermore, for a given substance, the exposure of an organism to it may vary according to the season, or even the time of day, in relation to: the manner in which the PPP is used (in particular the seasonality of treatments); the variability of the dominant transfer processes; and the variability of the life stages and behaviour of the organism. These parameters are indeed conditioned by different environmental factors such as temperature, light or humidity. For example, the presence of certain pollinators in a flowering plot, and therefore the exposure to PPP that may result, is strongly determined by the season, the time of day and the temperature. Similarly, in the marine environment, the accumulation of PPPs (in this case POPs) in right whales (and their consequent effects) are influenced by the variability of prey during migration cycles, or the mobilisation of lipids during periods of low feeding (Weisbrod *et al.*, 2000).

Finally, in agricultural areas, the type of crop affects both the presence of certain organisms (which may be preferentially present in orchards, or in cereal crops, etc.) and that of certain substances whose use is geared to the type of crop.

### Measures for limiting contamination and exposure

Various actions to limit and manage environmental contamination by PPPs were already identified during the previous CSA (Aubertot *et al.*, 2005b). These measures mainly consist of limiting the use and dispersion of PPPs at the time of application and reducing

post-application transfers at the plot and the broader landscape scales. Work on buffer zones, which form part of the broader landscape approach, are more recent than work at the plot scale. The management of effluents (e.g. tank bottoms) also represents a measure to limit point source pollution by PPPs.

The literature analysing the effectiveness of these measures focuses mainly on improving the parameters of the practices and devices implemented at these different scales, and on managing their complementarity. Some work also focuses on remediation strategies, consisting either of stimulating the natural degradation processes of substances, or of introducing artificial pollution treatment. Whether using a strategy of limiting transfers or by remediation, it is generally agreed that no measure can completely prevent the dispersion of compounds between environmental compartments and the consequent exposure of non-target organisms within them.

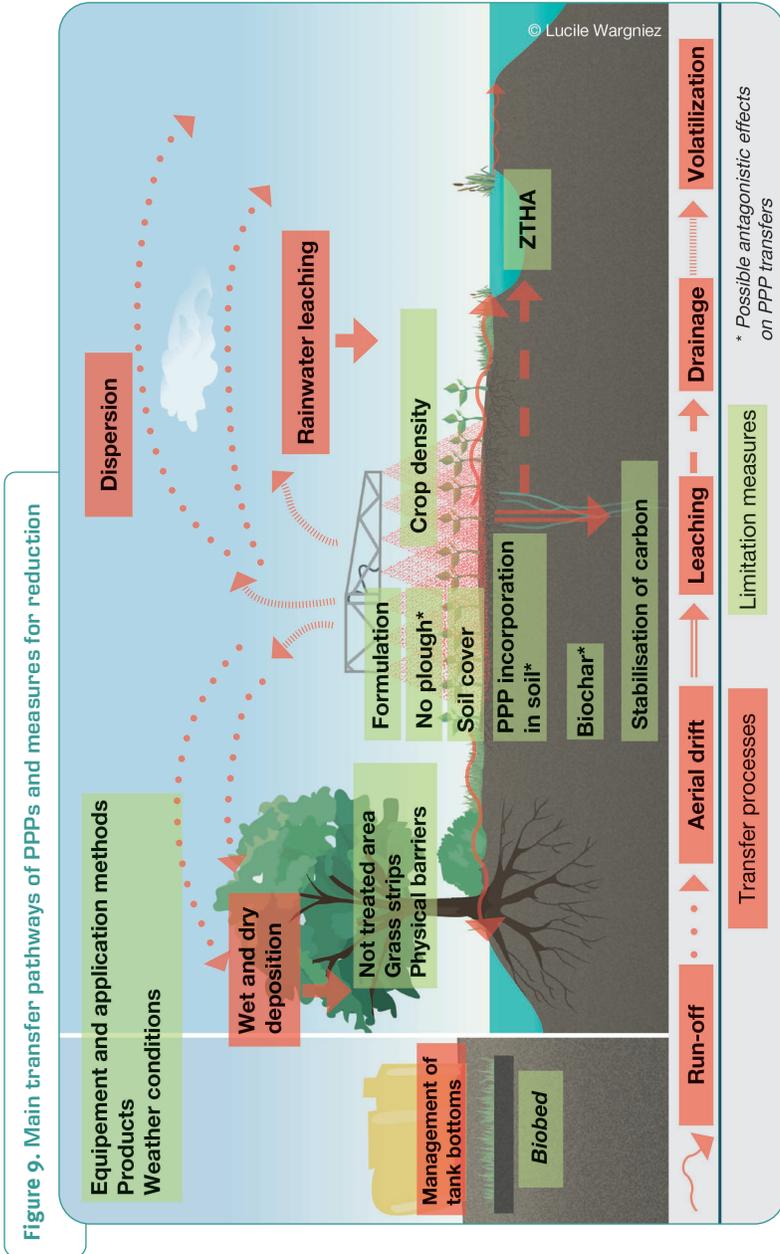
## ■ Limiting transfers to the environment

The main strategies for limiting transfers are based on the retention of substances (in plant biomass, soil or sediment) and their degradation. Both processes can have indirect effects. Retention is generally only transient and can lead to a delay in the transfer and/or bioavailability of substances and exposure of organisms. Degradation reduces the concentration of the substance present, but may increase the concentration of transformation products. Finally, the retention and degradation processes may be antagonistic. The entrapment of substances can, in effect, lead to their removal from the metabolic activity of microorganisms, thus illustrating the need to find a compromise.

Since there are multiple transfer pathways for PPPs, limiting one may sometimes favour another. For example, certain devices limiting horizontal transfers through runoff can favour vertical transfers through leaching and the entrainment of substances into groundwater. The spatial positioning of installations thus requires consideration of all parameters (Figure 9).

Another conflict exists between limiting the exposure of organisms and the effectiveness of application methods, i.e. between protecting non-target organisms and the desired exposure of target organisms. Indeed, some measures that protect biodiversity tend to limit the effectiveness of the treatment. It has been observed, for example, that the presence of mulch can limit the contact of herbicides with the soil, leading to an increase in doses needed to obtain the desired result. Furthermore, some adjuvants used to improve the contact of the product with its target, and thus reduce the required rates, may at the same time increase the propensity of the substance to be transferred to the environment.

Limiting losses to the environment must be thought through in a coherent way across the different scales, and concerns the management of PPP application and tank emptying, the management of the plot, and the planning of the surrounding landscape (Figure 9).



## During application

The main sources of transfer identified during application are drift and volatilization, where the dose can condition the intensity of transfer.

Reducing the quantities applied is the first means of reducing environmental exposure to PPPs. The possible strategies for bringing about such a paradigm shift have been the subject of studies conducted in parallel with this CSA (in particular the CSA on natural pest regulation and the priority research programme 'Growing and Protecting Crops Differently' mentioned in the introduction).

The choice of product (composition, formulation) will have an impact on drift and volatilization. The formulations contain different adjuvants and co-formulants in different concentrations. These are intended to reduce drift, but they also have functions of wetting, spreading, adhesion, retention and resistance to wash-off, especially on plant leaves with hydrophobic properties. However, the addition of adjuvants is less effective than the use of low-drift nozzles. On the other hand, an adjuvant that modifies the behaviour of the active ingredient, for example by promoting the penetration of the PPP into the plant, should reduce volatilisation, but few studies have been conducted on this subject. Finally, these modifications to the behaviour of the active ingredient may also have consequences for its bioavailability and therefore for the exposure of non-target organisms.

Among the most recent formulations, nanopesticides cover a wide variety of products that combine several surfactants, polymers and nanoparticles in the nanometer size range. These nanoformulations improve the apparent solubility of poorly soluble active substances, as well as their gradual release and/or protection against premature degradation. They thus reduce the dosage of PPPs, but they can also create problems related to more efficient transport and greater persistence in soils, waters and organisms. Furthermore, there is little research to date on the overall assessment of the fate of nanoformulation coatings in soil and the environment after release of the active substances, nor on their redistribution in plants after absorption, and no studies on environmental exposure.

Replacing spraying with alternative processes such as seed treatment eliminates the risk of spray drift, but is likely to generate aerial transfers of PPPs in the form of particles. While the deflectors made compulsory on pneumatic seed drills since the order of 13 April 2010 appear to be effective in reducing concentrations in the air, deposits and emissions of the coarsest dust downstream of the treated plot, or the emissions themselves, they are less effective for fine (micrometric) particles and generate a cloud of soil dust. Limiting the dispersion of PPPs at the time of sowing can also be based on improving the seed treatment itself (adhesion and applied dose). However, as will be discussed later, the use of treated seed can cause contamination in granivorous animals.

The timing of application has consequences for transfer and exposure, which depend on weather conditions and the behavioural traits, phenology or life stage of non-target organisms present in the agroecosystem, in combination with environmental parameters. Meteorological conditions (wind, precipitation, humidity and temperature, soil moisture)

are crucial for the risk of transfer of PPPs, especially through runoff, drift or volatilisation. They also have a direct influence on the deposition of PPPs and therefore on their biological effectiveness. The most favourable conditions correspond to low winds (spraying is prohibited in France from a wind speed of 3 on the Beaufort scale, i.e. 19 km/h at a height of 10 m), a moderate temperature and average humidity to limit the evaporation of drops, a soil that is neither too dry to allow the product to be distributed between the soil solution and the solid matrix, nor too wet to avoid runoff (the application of PPPs is prohibited in France when the intensity of rainfall is greater than 8 mm/h). Finally, as the occurrence of rainfall after application plays a crucial role in the risk of transfer, an important strategy is to avoid treatments before rainfall.

However, meeting these conditions is not always possible for farmers, and it can sometimes conflict with the objective of crop protection, especially in the case of fungal disease treatment where rain can facilitate spread from the soil.

The choice of application equipment is a commonly identified means of limiting losses through drift. The factors that can be controlled include the size of the spray droplets, the management of air assistance (co-flow), the containment of sprays, the porosity of the vegetation, etc. The average number of applications, the application equipment used and the architecture of the vegetation are also significant factors. Much work is being carried out on the development of innovations in this regard (e.g. low-drift nozzles, air flow devices, and recovery or containment panels), the effectiveness of which remains dependent on the conditions of use.

### When emptying tanks

Poor management of PPP effluents (tank bottoms) can contribute to significant risks of transferring these substances, via point source pollution processes that are otherwise easily controlled. In order to prevent these risks, the decree of 4 May 2017 on the sale and use of PPPs provides for a list of effective treatment processes for plant protection effluents, established and published by the Ministry of Ecological Transition<sup>16</sup>. The scientific literature is more extensive with regard to the ability of biobeds to significantly reduce contamination from the washing of treatment equipment and the management of tank bottoms. A biobed consists of a pit filled with a substrate capable of retaining the PPPs contained in the tank rinsing liquid that has been poured into it. These substances decompose due to the enzymatic degradation power of the microorganisms present in this substrate, particularly that of fungi. Thus, biobeds activate complex mechanisms that combine the stimulation of metabolic activity with sorption processes. Two very important factors must be considered for the proper functioning of these biobeds: the composition of the substrate (biomix), which must be pre-composted and locally assessed as a function of the materials used along with the PPPs to be degraded, and the management of the humidity of the biomix, which must favour microbial activity. The maturation time of

16. [https://www.bulletin-officiel.developpement-durable.gouv.fr/documents/Bulletinofficiel-0030426/met\\_20180009\\_0000\\_0022.pdf;jsessionid=6E916A22B2D12A59C43CBBE6EDE294DB](https://www.bulletin-officiel.developpement-durable.gouv.fr/documents/Bulletinofficiel-0030426/met_20180009_0000_0022.pdf;jsessionid=6E916A22B2D12A59C43CBBE6EDE294DB) (accessed 9/01/2023).

the biobed ranges from one to eight months, with contrasting effectiveness depending on the molecules. The biomix is then reapplied to the plots, although no studies have yet thoroughly characterised the nature of its impacts on biodiversity and the functions of soil (micro)organisms. On the other hand, PPP losses by volatilisation during this re-application may occur.

### At the level of plot management

Some agricultural practices influence plot characteristics that are crucial for transfer, such as soil cover, soil structure or organic matter content. These factors mainly influence the fate of PPPs (adsorption, degradation, storage), exports from the soil through runoff (erosive or not) and infiltration into the profile (leaching), as well as those through post-application volatilization or wind erosion into the atmosphere. The management of the soil component, which is one of the main filters for reducing PPP transfers, thus represents a primary control measure.

Soil cover (e.g. presence of a cultivated plant cover, mulch of natural or non-natural origin, grass cover) directly interferes with the transfer of PPPs: it allows them to be intercepted by the foliage, thus delaying their arrival in the soil. However, the trapping of PPPs in plant tissues (crops and/or weeds) hinders their degradation by microorganisms, which increases their persistence. Furthermore, when crops (or weeds) senesce, the trapped PPPs can be released back into the environment if they have not been degraded. Soil cover also limits surface runoff, although the ability of mulches to limit leaching of PPPs is controversial: maintaining high soil moisture may contribute to vertical movement of PPPs. The presence of a mulch is also likely to favour the volatilisation of PPPs by increasing the exchange surface with the atmosphere, by modifying the temperature and humidity conditions and by modifying the availability of the product (adsorption or non-adsorption onto the mulch, degradation). However, the effects of mulch on cumulative volatilization losses are not yet well known. Finally, the cover can play a more indirect role on the soil structure and the activity of microorganisms, favouring the degradation of substances.

Conversely, when the cover is non-permeable (plastic mulch or pavement in urban areas and infrastructures), transfer by runoff is aggravated.

The addition of biochar, developed in recent years in order to store carbon in soils and combat global warming, also affects the adsorption processes of PPPs. Biochars are carbonaceous substances obtained by pyrolysis of biomass in an oxygen-limited atmosphere. They have the particular characteristic of being resistant to degradation, and can have a sorption capacity two to three times greater than that of the soil. Research by Blanco-Canqui (2019) and Khorram *et al.* (2016) shows that by promoting the trapping of PPPs in biochars, they are less likely to be leached, and that by improving the physical properties of the soil surface (porosity, water retention), biochars strongly reduce erosive processes and thus the transfer of PPPs by erosive runoff. Biochars may also be applied to sequester PPP residues in contaminated soils and to reduce uptake by plants. However, studies have shown variable effects of biochars on PPP sorption depending on the input material and particle size of the biochars, the time elapsed after application,

the application rate and the pyrolysis process used. Furthermore, one drawback identified by Yavari *et al.* (2015) is the reduced efficacy of PPPs on targets (weeds, fungi) when applied to soil treated with biochars. Ultimately, field studies are still needed to investigate the effects of biochars on the transfer of PPPs in field conditions.

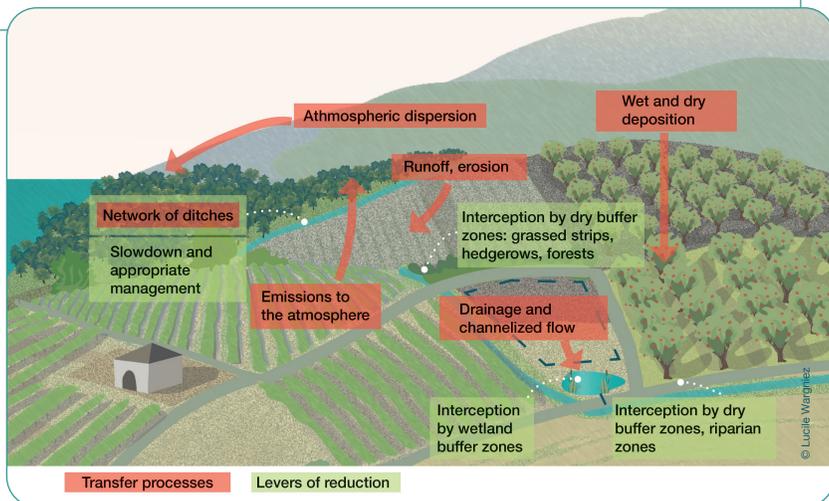
Tillage leads, more or less temporarily, to the modification of soil properties, thus affecting the fate of applied PPPs, notably promoting runoff transfers. Reducing tillage helps to increase surface organic matter, increase microbial activity and stabilise pH and moisture levels; these conditions can facilitate the interception, retention and degradation of PPPs. This type of practice limits erosion and therefore the transfer of PPPs through surface runoff. However, the absence of tillage can, on the other hand, lead to the emergence of preferential flows (macroporosity) that favour the leaching of PPPs. On the other hand, by allowing the incorporation of PPPs into the soil, tillage limits their volatilisation. In conclusion, whether or not the soil is tilled leads to risks of PPP transfers even if the transfer routes are different.

The movement of agricultural machinery around fields can lead to soil compaction, which favours erosion, runoff and the transfer of PPPs adsorbed onto soil particles. It is therefore a matter of reducing mechanised interventions and considering the choice of machinery, as well as organising permanent passageways, also known as *controlled traffic farming* (CTF), for which remote control tools on board the tractors are proposed. Decompaction (*loosening*) of soil or early application of PPPs in localised strip treatments, combined with controlled traffic farming, also make it possible to limit the risks of PPP transfer by runoff.

Water management, whether in terms of irrigation or drainage, has important consequences for transfers. Inappropriate irrigation practices, or those carried out during a high-risk period, can promote runoff and leaching of substances. It is therefore necessary to limit irrigation practices, especially on mulch, when aiming to promote the penetration into the soil of herbicides used as pre-emergence treatment for weeds. Agricultural drainage is a technique to evacuate excess winter water from hydromorphic soils. It is recognised that losses of PPP through drainage systems, although appreciable, are on average less than losses due to runoff and erosion, but greater than losses due to leaching into aquifers. Thus, most measures to mitigate leaching losses (notably tillage) will also reduce drainage losses, as will the measures recommended to mitigate runoff and erosion losses. However, the main way to avoid PPP losses through drainage is to restrict the period of application to the period when drainage is not active, and to take into account the moisture content of the soil: the drier the soil, the lower the vertical transfer. The Soil Wetness Index (SWI, produced by Météo-France) could be used to schedule PPP applications based on the water content of drained soil profiles.

### At the landscape level

In addition to actions to limit the transfer of PPPs within agricultural plots, certain landscape designs or natural landscape features can play an important role in mitigating PPP transfer and exposure of non-target organisms (Figure 10).

**Figure 10. PPP transfer at the catchment scale and measures to reduce it**


Buffer zones (BZs) can be divided into dry buffer zones (DBZs), such as grassed strips, hedgerows or copses, and wet buffer zones (WBZs), whether natural, such as marshes, mangroves and lagoons, or man-made (WBZs constructed to mimic natural wetland conditions and processes). Properly constructed ditches or drainage courses can also play an intermediate role between these two types of BZ.

BZs act as an interface between PPP application sites and the surrounding environment (watercourses, housing). They also function as a reservoir of biodiversity and refuge areas for organisms, and play a major role in the recovery process between successive applications. In some cases, however, accumulations of PPPs and transformation products have been found at the foot of hedges and in lentic systems (still water bodies), creating a significant risk for the biodiversity present in these environments.

DBZs can mitigate flows and concentrations of PPPs in runoff, particularly by promoting infiltration, but also sedimentation, adsorption and dilution. Vegetation plays an important role not only in its ability to intercept runoff due to the slowing of surface flows, but also in its ability to adsorb PPPs on the surface of the DBZ or in the root zone. These horizons have a high organic matter content which favours the retention of PPPs and reduces their leaching. However, measurements have shown a rapid appearance of transformation products in the surface horizons of DBZs, associated with the formation of bound residues that may be released in the longer term.

The size and positioning parameters should be considered in relation to the volumes of incoming runoff, the infiltration capacity of the DBZ, the residence time of the water

and the adsorption capacity of the DBZ. For example, the BUVARD tool (Catalogne *et al.*, 2018) could be used to help identify the required grassed strip width for the desired runoff abatement efficiency. Thus, the grassed strip width factor, while important, is not sufficient in itself to ensure good transfer mitigation.

The location of the grassed strips within the catchment area is also a determining factor in their effectiveness. They should be installed sufficiently far up the slope to limit the surface area contributing to runoff and the risk of runoff concentration. Conversely, positioning them at the bottom of the slope can lead to problems, as there is a greater risk of runoff concentration in gullies. Furthermore, the proximity to the watercourse increases the risk of water logging, which limits infiltration and increases the risk of contamination of the shallow water table. In addition, it is essential to minimise short-circuiting flow and to avoid soil compaction or saturation within the DBZ, which greatly limits its effectiveness. The siting must therefore be the subject of a hydrological diagnosis on the scale of the catchment area as well as the installation site itself.

This study shows the importance, already highlighted in the previous CSA addressing this issue (Aubertot *et al.*, 2005b), of encouraging research that combines the monitoring of both parent molecules and transformation products under natural conditions. On the other hand, the risk of vertical transfer and remobilisation of PPPs and their transformation products, over time, into groundwater remains insufficiently studied. Finally, the bibliography highlights the difficulty of measuring, in the field, the effects of the establishment of DBZs at the catchment scale on water quality. New tools for measuring PPP concentrations and flows, such as passive integrative samplers, can help progress this research if they are supplemented by appropriate hydrometeorological monitoring and sufficiently detailed knowledge of the actions actually implemented by farmers at the catchment scale.

As regards aerial transfers, any device increasing the distance and/or representing a barrier between the edge of the treated plot and the ecosystem to be protected can reduce these transfers. In particular, vegetation hedges represent natural physical barriers to reduce the atmospheric dispersion of PPPs, but artificial vertical systems, such as windbreaks or *Alt'Dérive*-style nets, can also be used to filter the air mass by intercepting droplets and modifying the airflow by decreasing the wind speed.

The effectiveness of physical barriers in limiting the dispersion of PPPs downstream of treated plots appears to be confirmed by measurements and modelling studies, which allow the integration of different factors in order to identify optimal values for the characteristics of these barriers (height, width, porosity, location). However, these characteristics are highly dependent on local parameters (soil, weather, topography, vegetation, gullies or ditches that may circumvent the DBZs) as well as on the sizing and positioning of these elements in the catchment area and with respect to the PPP application sites. This effectiveness must be evaluated, for both sedimentary and aerial drift. A combination of measures can also improve drift reduction.

WBZs are likely to intercept channelised water, either from channelled runoff via ditches in particular, or from agricultural drainage, in order to control PPP flows that occur in runoff. The most significant processes for reducing PPP transfer are, in decreasing order of importance, sedimentation, sorption, microbial degradation, photolysis, hydrolysis and uptake by vegetation. The latter is involved in three different mechanisms:

- direct absorption and accumulation of PPPs in plant tissues;
- production by the root system of enzymes that promote biodegradation;
- the combined effect of the vegetation and the microorganisms in the rhizosphere (phytostimulation can increase the activity of the microorganisms by five to ten times).

Hydraulic residence time, which is related to the hydrological response and dependent on buffer zone size, also plays an important role in the fate of PPPs: it takes approximately one month to significantly increase the dissipation of molecules (Stehle *et al.*, 2016). Finally, the performance of WBZs also depends on the season.

Factors that can be controlled in the design of WBZs include sizing (ratio of buffer area to the connected upstream hydrological area), the vegetation cover, the amount of organic matter, and the presence of substrates that support microorganisms. Recommendations converge towards a size greater than 1% of the connected upstream watershed (Tournebise *et al.*, 2017). Thus, to optimise a wetland buffer area and to maximise PPP/substrate contact areas, it is recommended that buffer zones are wide. This helps to reduce flow velocities, favours shallower areas (< 50 cm) and thus facilitates the establishment of aquatic vegetation and the processes of sorption and degradation.

Several reviews report WBZ efficiency values above 80% for the majority of PPPs (those that tend to be highly adsorbed), but below 40% for others (Stehle *et al.*, 2011; Vymazal and Bfezinova, 2015). In some cases, negative efficiency has been observed, resulting from PPP release processes due to remobilisation during strong flood events and/or desorption from sediments in the case of weakly adsorbed molecules.

The analysis of the literature for different types of WBZs showed that ponds have a significant effect in reducing the average concentrations and maximum PPP peaks between their inlets and outlets (between 60 and 100%). However, retention or degradation processes are often difficult to demonstrate because of strong dilution effect of the large water volumes in ponds (Le Cor *et al.*, 2021). Mangroves (coastal ecosystems at the interface between land and sea) provide conditions for remediation via PPP uptake by vegetation, accumulation, detoxification, retention and degradation (Ivorra *et al.*, 2021). In addition, hydrological conditions can favour these processes by increasing sedimentation and slowing down runoff. Rice fields have an efficiency ranging from 26% to 75%, as the flooded conditions enable the interception of irrigation water with varying PPP loads (Matamoros *et al.*, 2020).

Among the artificial WBZs (AWBZs), peri-urban ponds play a buffer role in the storage of PPPs, leading to a significant risk for the biodiversity present. The presence of vegetated strips (> 2 m) around these ponds would reduce the presence of PPPs (Ulrich *et al.*, 2018). Stormwater ponds, which are AWBZs designed to manage stormwater (flood

risk, water quality), are also highly effective in dissipating PPPs (36-100%, Cryder *et al.*, 2021). Maintenance and regular cleaning of these AWBZs allows the renewal of the sediment matrix, which contributes to the storage of hydrophobic molecules, thus raising the question of sediment management according to their contamination and the associated risk. Other solutions for intercepting agricultural flows through installations (e.g. flooded riparian strips, bioreactors; Tournebize *et al.*, 2020) have been evaluated for the retention of nitrate ions, but very little work has been done on their application to the case of PPPs.

The landscape emerges as a potential mechanism to limit the transfer of compounds into the environment, by modifying practices according to the vulnerabilities of the different zones within the landscape, by adjusting the spatio-temporal organisation of the plots (combination of use and practices) and the agro-ecological diversity in order to aim for an overall reduction in the use of PPPs and to increase the resilience of the landscapes to transfers (and to impacts). Various ongoing projects should shed new light on the tools that can be used at this scale.

### Modelling transfers

Given the multiplicity of PPPs, agro-pedoclimatic contexts and organisms, it is impossible to carry out laboratory experiments and field monitoring to assess the fate and impacts of all PPPs in all environments and for all biodiversity. Moreover, the processes involved are complex, non-linear and variable in time and space. Modelling therefore appears to be an essential tool for formalising, integrating and prioritising all the processes. To date, the models developed to simulate the fate of PPPs in the environment do not precisely reproduce the reality of transfers due to the complexity of the processes to be integrated. On the other hand, they do make it possible to compare risk situations, to define potential exposure levels or predicted concentrations in the environment for risk assessment and management. They also help to establish and test agro-pedoclimatic scenarios with the aim of reducing PPP transfers and associated risks.

The models in the literature are designed at different scales. At the plot scale, they simulate the transport of water and PPPs in the soil and their transfer to different environments (groundwater, surface water, plants, air). Other models developed at a local scale focus on evaluating the capacity of BZs to mitigate transfers based on different parameters such as strip width, roughness and vegetation density. Complex models have been developed for ditches (e.g. TOXSWA, TOXic substances in Surface WATers) and grassed strips (e.g. VFSSMOD, Vegetative Filter Strip Modeling System). VFSSMOD, for example, has been tested successfully, showing close agreement between the model predictions and the efficiency of PPP trapping by vegetation. However, further studies are needed to better account for the interactions between PPPs, soil and vegetation as they pass through BZs, as well as colloidal transport, preferential flow, retention and remobilisation of PPPs in the long term. For other types of buffer zones (hedgerows, WBZs), further studies are needed, as no model of PPP water transfer has yet been identified in the literature.

At the catchment scale, there are simple approaches based on Geographic Information Systems (GIS) and simple equations or scores based on expert opinion to determine the transfer and

mitigation potential of PPPs. These methods can be used as a first step to help identify risk areas within a territory. However, their performance is not always tested and they do not incorporate the temporal variability of the processes involved. There are many models at the catchment scale (e.g. I-Phy-Bvci, LEACHM-runoff, MHYDAS, PESHMELBA, SACADEAU, SWAT), but not all of these incorporate the influence of BZs. Among them, the SWAT model (Soil and Water Assessment Tool), which simulates the presence of BZs (e.g. grassed drainage courses, grassed strips, sedimentation basins), is the most widely used model at the international level. However, the spatial heterogeneity of the landscape elements (size, soil type, nature and density of vegetation, gradient), their actual location and their hydrological connections with the treated plots are not sufficiently well represented. These models are primarily research tools that are constantly being developed and improved. They offer interesting perspectives for determining the effectiveness of different combinations of buffer infrastructure. Finally, the conversion of these catchment-scale models into practical tools is still a challenge. Further, the ongoing development of tools coupling hydrological and atmospheric pathways at the scale of the catchment area should make it possible to estimate the various exposure pathways of non-targeted ecosystems within the catchment area.

As regards emissions to the atmosphere, models fall into two main categories, those that describe the processes involved at the time of PPP application (e.g. AgDRIFT, IDEFICS, DriftX), and those that describe the processes involved after application. The latter models predict volatilization from a treated plot by describing emissions from the soil and plant cover (e.g. Volt'Air-Pesticides, SurfAtm-Pesticides). Some models include the dispersion of the gas phase downstream of the treated plot, as it may generate exposure to PPPs via surface deposition (e.g. EVA 2.0, FIDES).

These models are diverse in their approaches, with complementary advantages and limitations. In general, analysis of these studies shows that the performance of these models could be improved by conducting studies to better document the representation of the effects of cultivation practices on PPP transfers, particularly with regard to innovative agroecological practices and the use of biocontrol. Certain processes relating to aerial transfers are also still insufficiently known, such as those concerning:

- spray droplet drift at the time of application or droplet characteristics;
- estimation of spray interception by the treated vegetation cover;
- the link between sedimentary and aerial drift or more systematic consideration of atmospheric stability conditions;
- post-application volatilization;
- the adsorption of PPPs from the gas phase to the solid matrix of the soil in the event of soil drying, or the interactions of the compound with the leaves (penetration, adsorption, photodegradation, leaching by rain);
- the effects of the formulation or of adjuvants on these processes.

The need to develop the expertise required to implement these models in the field and to take better account of the uncertainties associated with the results of the models are also highlighted in the literature.

## I Remediation

Remediation of PPP-contaminated environments is sometimes possible, but never complete. A combination of biotic and abiotic methods can be used to promote PPP degradation processes (Fenner *et al.*, 2013).

When an area is contaminated by PPPs (soil in particular), biotic remediation (bioremediation, phytoremediation, rhizoremediation) represents a cost-effective, non-invasive and acceptable means of eliminating the substances (Arthur *et al.*, 2005). It generally involves the use of plant cover and the inhibition/stimulation of microbial biodegradation capacities, i.e. the partial or complete conversion of the PPP into its elementary constituents (Megharaj *et al.*, 2011). Rhizoremediation, within the rhizosphere, and phytoremediation, involving plants usually in association with symbiotic microorganisms (e.g. rhizobacteria and mycorrhizae), also allow for the metabolisation and degradation of PPPs (Eevers *et al.*, 2017). The bioavailability of a PPP is the main limitation for effective phytoremediation. Some PPPs may be resistant to degradation and/or be toxic to plants and microorganisms that do not possess the appropriate enzymes (Eevers *et al.*, 2017). Furthermore, in the case of phytoremediation, plants must be collected and incinerated or composted to dispose of the PPPs. Finally, few studies have been carried out under real conditions to determine the effectiveness of these techniques in reducing PPP transfers.

To reduce the exposure of organisms, abiotic remediation methods can be implemented *in situ* (use of surfactants to promote leaching of PPPs, vitrification, separation, containment by physical barriers) or *ex situ* (excavation, thermal treatment, chemical extraction, encapsulation). They are generally expensive, given the diffuse nature of PPP contamination, and they raise issues about the consequences of these processes on the structure and properties of soils.

Given the absence of regulatory obligations to restore environments, the various existing remediation methods are not generally implemented.

## New developments and prospects for characterizing contamination and exposure

Improving knowledge of the degree of contamination of the environment by PPPs depends essentially on a combination of the latest sampling strategies and analytical techniques to provide a more complete view of the situation, including its dynamic aspects. The difficulty in identifying reliable medium- and long-term contamination trends in the various environments highlights the need for national monitoring plans that are standardized in terms of frequency, sampled matrices and methodologies (from sampling to analysis, and even data processing) in order to better respond to the needs and issues at play; see, for example, Hulin *et al.* (2021) in relation to the atmosphere. The factors that determine the bioavailability of PPPs are not yet sufficiently known to allow specific sampling of the bioavailable fractions.

However, research on the contamination of biota has shown the importance of impregnation of organisms and transfers of substances within food webs. Finally, modelling is included for the answers it can provide to the need to combine different scales and test scenarios.

## ■ Developments in sampling strategies

Evolving sampling strategies are contributing to a steady increase in the number and diversity of substances analysed in the different environmental matrices.

In order to characterise trends in contamination over time or to identify low concentrations of a chronic nature in the environment, one of the most important developments since the beginning of the 2000s concerns the development and implementation of passive integrative samplers. These allow for an improvement in the representativeness of measured chronic contamination levels (by integrating a period of several days to several weeks of exposure) and for the quantification of certain substances that are not detectable on the basis of one-off samples. Table 1 shows the most commonly used passive integrative samplers in the aquatic environment. To date, only DGT (diffusive gradient in thin film) and POCIS (polar organic chemical integrative sampler) have been used since 2012 for WFD monitoring of hydrophilic PPPs in Mediterranean coastal waters, but the use of the various existing passive integrative samplers makes it possible to meet several analytical challenges in order to improve the mapping of environmental contamination (Bernard *et al.*, 2019). It should also be noted that developments are underway for passive integrative samplers for the atmosphere (Galon *et al.*, 2021).

**Table 1. The most commonly used passive integrative samplers for PPP sampling in the aquatic environment**

Passive integrative sampler	Types of PPPs sampled	Regulatory use
DGT ( <i>diffusive gradient in thin film</i> )	Inorganics and hydrophilic organics	WFD
POCIS ( <i>polar organic chemical integrative sampler</i> )	Medium-polar and polar organics	WFD
Chemcatcher®	Medium-polar and polar organics	
SPMD ( <i>semi-permeable membrane device</i> )	Hydrophobic organics	
LDPE ( <i>low density polyethylene</i> )	Hydrophobic organics	
MESCO ( <i>membrane-enclosed sorptive coating</i> )	Hydrophobic organics	

The analysis of PPPs in biota can also provide an integrated view of contamination and exposure of organisms. However, it is important in this type of approach to take into account various criteria that condition the exposure and accumulation of PPPs in organisms under

real conditions (*in natura*). These criteria include the chemical nature of the substances in question, the temporal variability of the location of the organisms and their developmental stage, differences in their sex, the type of organism in question and its metabolic capacity, as well as the variability of the different contamination pathways in the biota (e.g. via respiration, the integument and/or food). The choice of taxa and the matrix used for this monitoring is therefore an important element to consider with regard to the various parameters mentioned above. For example, the use of moult feathers makes it possible to record exposure to PPPs in birds over several weeks or months, whereas the passage of these substances through the bloodstream is brief. The development of alternative biomonitoring approaches by holding organisms in cages *in situ*, applied in particular in aquatic environments, may make it possible to partially overcome these constraints. This is the case, for example, of *in situ* encagement of gammarids for measuring the bioaccumulation of chemical substances (e.g. Afnor standard XP T90-721, 2019).

Furthermore, to develop a more exhaustive understanding of PPP contamination and its transfer between matrices and across environmental continuums would require more consideration of the spatial aspect of the sampling strategy, both on a large scale (by distributing monitoring over the entire national territory, including the overseas territories) and on a more local scale (by considering the connectivity of environments and the different matrices that make them up). Similarly, it would be relevant, among other things, to broaden the range of these matrices in monitoring and scientific studies (e.g. rainwater, Potter and Coffin, 2017; deep marine areas, Munsch *et al.*, 2019). It would also be useful to improve understanding of the fate of PPPs, by taking better account of the formation of transformation products and by coupling the acquisition of relevant data sets with the development of models aimed at better predicting the spatio-temporal behaviour of PPPs in the different compartments. These models, which are currently being developed and tested, could notably be used to refine spatio-temporal monitoring strategies.

## ■ Evolution of analytical techniques

The evolution of analytical techniques is another factor contributing to the increase in the number and diversity of substances analysed, but also to the improvement in the sensitivity of measurements. Recent years have seen the development and application of new and improved methods, making it possible to consider a greater number of chemicals simultaneously (including certain transformation products) and, to an extent, to dispense with the need to make *a priori* choices (e.g. multi-residue analyses or non-targeted analyses).

It is indeed necessary to have a more complete picture of the contamination by combining the available analytical technologies (gas chromatography [GC] and liquid chromatography [LC] coupled with tandem mass spectrometry [MS/MS], for sensitivity, with potentially less need for upstream extraction/preconcentration stages, particularly for water) with high-resolution mass spectrometry (HRMS). The latter enables a wider range of detectable and identifiable substances to be found, initially using screening approaches for suspected substances, and subsequently using non-targeted screening approaches, i.e. without any

*a priori* choice of substances to be sought (Gonzalez-Gaya *et al.*, 2021). The adoption of these approaches would in particular make it possible to broaden the spectrum of transformation products searched for. However, these approaches will only be able to develop fully if new standards become available (particularly concerning transformation products) in order to definitively confirm the presence of PPPs not analysed to date (including co-formulants and adjuvants) and to determine their concentration levels. The development and implementation of these new analytical technologies, beyond specific academic studies, will also raise new challenges for managing and sharing the large amounts of data generated. The evolution of analytical techniques can also contribute to a better understanding of the effects of PPPs in food webs. For example, the analysis of the isotopic signatures of carbon, nitrogen, sulphur or even oxygen in different matrices can be used as a *proxy* (i.e. a substitute variable) to detect qualitative or quantitative changes in the structure of food webs caused by PPPs.

## I Characterizing exposure

Further research is also needed to better relate measured contamination levels to the resulting risks and impacts on organisms and ecosystems. In particular, this implies taking better account of the diversity of exposure pathways. The dynamics that determine the fate of substances could be better understood by considering the formation of transformation products and the possible interactions between the various compounds. The distinction between isomers (in particular enantiomers), which are spatial arrangements of molecules with the same physico-chemical properties but different biodegradation dynamics and physiological effects, also appears to be an important element for better explaining the relationship between contamination and effects. The adequacy of these measurements is essential for the development of models aimed at better predicting the spatio-temporal evolution of PPPs in the various compartments and the resulting exposure of organisms.

## I Theoretical assessment and modelling

As mentioned above, much research is being conducted on the development of models to simulate the fate of PPPs in the environment at the plot level (transport of water and PPPs in the soil and their transfer to other components: groundwater, surface water, plants, air, etc.) and at the watershed level, or to model the capacity of buffer zones to mitigate transfers. They allow potential exposure levels or predicted concentrations in environments to be defined in order to carry out risk assessments. They also help to establish and test agro-pedoclimatic scenarios with the aim of reducing PPP transfers and associated risks. In general, despite many recent developments, currently available models are not able to describe all the processes involved, nor the great diversity of existing agricultural practices, and no model integrates the land-sea continuum. The temporal evolution of PPPs in soils also remains difficult to predict. It is also necessary to work in parallel with field observations for the design, development and parameterisation of these models, by integrating, for example, data on the use of PPPs (e.g. surveys of farmers or public databases).

PPP fate models aimed at informing PPP exposure risks should also be linked to models that account for the ecology of organisms (e.g. probability of presence and/or movement of animals in the landscape matrix) based on direct observations, telemetry or GPS tracking of individuals, or habitat selection models.

Similarly, models that aim to assess bioaccumulation could be made more robust by incorporating new parameters that are important for the bioaccumulation and biomagnification of PPPs. Indeed, parameters such as the octanol/water partition coefficient ( $K_{ow}$ ) and the bioconcentration factor (BCF), which are typically used, have been selected mainly on the basis of studies of highly lipophilic substances with a high  $K_{ow}$ , which does not correspond to many PPPs currently in use, and from studies mainly concerning aquatic systems. Recent work indicates that absorbed organic substances that are not metabolised, or their transformation products, have the potential to bioaccumulate in air-breathing organisms, and to bioaccumulate (or even biomagnify) in terrestrial food webs when they have a  $\log K_{oa} \geq 5$  and a  $\log K_{ow} \geq 2$ . Conversely, substances with a  $\log K_{ow} < 2$  are generally eliminated rapidly by urinary excretion and therefore do not biomagnify, even if their  $\log K_{oa}$  is  $\geq 5$ .

Overall, the approaches for measuring PPP residues in organisms *in natura* now make it possible to learn about the actual mixtures to which organisms are exposed. This is particularly the case with data provided by the ERBFacility (European Raptor Biomonitoring Facility) network. ERBF biomonitors raptors using multi-residue approaches in order to propose a global, pan-European analysis of environmental contamination as reflected in the contamination of raptors. It takes into account the many biological and ecological features specific to the species monitored (diet, distribution area, habitat, migration), in relation to various human activities (including agricultural activity).



# 3. Effects on biodiversity

In this CSA, biodiversity is addressed in its genetic, taxonomic (species) and functional dimensions. Taxonomic diversity is mainly described by the species richness (number of species), the relative abundance of species within the communities they comprise and the degree of heterogeneity of the latter. Within each species, genetic diversity is examined (intraspecific diversity). This can be described by phylogenetic metrics and population genetic indices. The degree of phylogenetic biodiversity is also assessed through indices of species richness and potential capacity to ensure a diversity of functions. The functional dimension can thus be described by identifying groups of species with similar functional characteristics and observing the number and diversity of these groups in a community. It is also important to note that each category of biodiversity can be estimated at different scales, from the local level (alpha diversity) to the regional level (gamma diversity), and in terms of dissimilarity between ecosystems (beta diversity). Initially focused on species diversity, this approach is now also applied to other components of biodiversity, and has led to recent developments that use unified concepts to estimate taxonomic, functional and phylogenetic biodiversity at different scales.

The effects of PPPs on biodiversity result from exposure to one or more substances, their toxicity, the stress resulting from the repetition and intensity of exposure, and the degree and distribution of exposure among exposed species, etc. They have indirect consequences due to the interdependence of species and manifest themselves at different levels of biological organisation, on different time and space scales, with different levels of severity and reversibility, which may in part depend on the characteristics of the exposure scenario. The effects on biodiversity status influence the ecological processes in which organisms are involved, and consequently the functioning of the ecosystem.

Due to the complexity of these interactions, the environmental context not only influences the relationship between contamination and exposure (see section on ‘Influence of context on exposure dynamics’), but also the relationship between exposure and effects. The factors influencing these dynamics are therefore presented in the section below. The main concepts commonly associated with the different types of effects are then reviewed to clarify their scope and limitations. The main effects highlighted in the analyzed bibliographic corpus are then summarized, distinguishing between effects on the status of biodiversity and those on ecosystem functions as defined in the CSA analysis framework. Finally, the main developments underway and the prospects for improving knowledge of the effects are identified.

## From exposure to effects, sources of variability in sensitivity to PPPs

The effects of PPPs on organisms depend firstly, as seen above, on the toxic properties of the substances and the conditions of exposure to these substances. However, depending on their biological characteristics, different organisms or groups of organisms subjected to the same exposure in the same environment will respond differently. This variability in sensitivity depends on characteristics that may be related to the exposure pathway and to certain biological traits (or biological characteristics) at the individual, species, population or community level. More generally, all of the factors that affect the status of organisms or groups of organisms (resource availability, threats, pressures and other contamination) will have a cumulative effect on their vulnerability (see section on ‘Contexts and situations leading to vulnerability’).

### I Influence of exposure pathways

Few studies have examined the influence of PPP exposure routes on observed effects. However, this influence has been demonstrated for different types of organisms (e.g. for hares, Mayer *et al.*, 2020, for birds, Mineau, 2002, or for trichoptera, Rasmussen *et al.*, 2017). Research on insecticides (organophosphates, neonicotinoids and carbamates) has, for example, led to the detection of residues of these PPPs on skin and/or feathers, highlighting the potential contribution of airborne sources to the exposure of wildlife via cutaneous and/or respiratory routes. The cutaneous route represents, for example, a significant exposure route for amphibians due to the thinness of their integument and their dual terrestrial and aquatic exposure, or for chiroptera due to the thinness of their wing skin. It therefore increases the total exposure and thus the associated risk. The cutaneous pathway of substances also modifies their metabolism by bypassing the liver, the main organ involved in detoxification processes. The question of exposure routes may also concern photosynthetic organisms, since certain differences in sensitivity to PPPs observed between macrophytes and planktonic microalgae may be explained by different exposure routes, i.e. via contact with water and/or sediments (e.g.: Vonk and Kraak, 2020).

Differences in toxicity depending on exposure pathways are difficult to generalise. They appear to depend on the routes themselves (e.g. exposure via the ambient environment or the trophic pathway), but also on the type of taxon and the substances considered. The few results on this subject suggest that these different pathways should be explicitly included in risk assessments for PPPs. This is important not only to better assess the actual exposure of an organism in its natural environment (and the potentially cumulative effects of the various possible pathways), but also to better take into account the effects resulting from trophic interactions.

### I Effects of accumulations and mixtures of substances

Exposure to PPPs may involve simultaneous exposure to different substances, or successive exposure (with varying degrees of frequency) to the same or different substances.

Thus, the accumulation and/or repetition of exposure of organisms to PPPs, whether identical or different, is a factor that influences the effects induced by these substances. For example, the impact of insecticides on arthropods varies with the number of treatments, as evidenced by a decrease in the abundances of spiders, carabid beetles, rove beetles, lacewings or ladybirds, following successive exposures to lambda-cyhalothrin (Wick and Freier, 2000; Liu *et al.*, 2013) or deltamethrin (Macfadyen and Zalucki, 2012). Karimi *et al.* (2021) also show that the ecotoxicological risk caused by copper depends not only on the dose applied, but also on the amount of copper already present in the soil. Under controlled conditions, repeated exposure to brodifacoum, a second-generation anticoagulant, has also been shown to induce more severe coagulopathies in the American kestrel than a single exposure (Rattner *et al.*, 2020). Similarly, repeated contamination with herbicides can induce aggravated effects on photosynthetic river biofilms (Tlili *et al.*, 2008). However, repeated exposure can also favour the selection of PPP-resistant species to the detriment of the most sensitive species, as demonstrated in heterotrophic and phototrophic microbial communities, particularly in the context of PICT (pollution-induced community tolerance) approaches. This factor also relates to the evolutionary processes of adaptation.

The question of the effects of PPP mixtures (sometimes referred to as 'cocktail' effects) has mainly been addressed by studies examining simultaneous exposure to different PPPs. The resulting results are highly variable and sometimes contradictory, depending on the combinations tested, the types of organisms studied and the experimental protocols used, which makes it difficult to identify robust trends. The experimental designs do not always allow for the demonstration of a genuine interaction between the PPPs, beyond a simple addition of the effects linked to the addition of the concentrations of each substance. The majority of studies describe an increase in the impacts of PPPs when organisms are exposed to PPP mixtures. However, some examples show potentiating or synergistic effects beyond the addition of the effects of each substance. In some cases, effects are observed for mixtures of substances, while at the same concentrations these substances have no effect when tested individually. Synergy between acaricides, used by beekeepers for hive maintenance, and fungicides potentially present in foraging areas, has been demonstrated for honeybees (Johnson *et al.*, 2013), and other examples are reported for the broader pollinator community in different contexts of exposure to several insecticides or to combinations of fungicides with other PPPs. A recent study assessing genotoxicity on onion in a germination bioassay shows that at low concentrations at which it has no effect alone, mesotrione in combination with atrazine shows a genotoxic effect (Felisbino *et al.*, 2018).

It is also important to note that studies dealing with the influence of adjuvants and co-formulants are relatively rare, except for glyphosate, for which polyethoxylated amine (POEA) adjuvants, acting as surfactants (POE-tallowamine), have sometimes been shown to be much more toxic than the active substance alone (Mesnage *et al.*, 2019). A few studies also document the increased toxicity of formulations compared to active molecules taken alone, such as the recent review by Nagy *et al.* (2020) which analyses the toxicity of 24 PPPs, mainly herbicides, and their formulations.

## I Distribution of levels of sensitivity and adaptation

The sensitivity of species to PPPs depends on a number of factors, including their proximity to the PPP target species, notably their phylogenetic proximity, which tends to make them sensitive to the same molecular mechanisms, their ecological proximity, or their way of life and habitat. Sensitivity can also vary within a species, between lineages, between populations, or even between individuals in the same population. When this variation has additive genetic elements, it can lead to evolutionary processes of adaptation involving natural selection.

Levels of susceptibility may vary over time due to a range of phenomena (e.g. acclimatisation, tolerance, resistance, and resilience) observed at different biological (individuals, populations, communities) and temporal scales (rapid and reversible physiological adaptation through developmental and phenotypic plasticity, or, on the contrary, longer-term adaptation through selective evolutionary processes that apply on a multigenerational scale). Indirect evolutionary effects can also affect susceptibility by reducing the genetic variability of populations and thus their adaptability to PPPs and other environmental pressures, especially when they increase the effects of genetic drift and inbreeding. This is the case, for example, when a PPP leads to population reduction, isolates populations from each other or favours a breeding system that reduces heterozygosity (through self-fertilisation or inbreeding).

The extent of evolutionary effects is probably largely underestimated to date, and they should be taken into greater account when assessing the effects and risks of PPPs. The ever-increasing number of cases of genetic resistance observed in species targeted by certain PPPs (insecticides, fungicides, bactericides) is a strong argument for taking such potential effects into account when considering the impact of PPPs on biodiversity.

For example, in aquatic invertebrates, the development of resistance to certain insecticides reflects an evolutionary process of selection (genetic adaptation) induced by the continuous or repeated exposure of populations. Such evolution has been observed in the freshwater crustacean *Hyalella azteca* species complex in response to the persistent presence of pyrethroids in sediments, favouring the development of different types of resistance involving the parallel evolution of different alternative mutations at the molecular target (sodium channel) previously identified in the target insects (Weston *et al.*, 2013; Major *et al.*, 2018). The development of resistance raises the question of the associated physiological or ecological cost. A recent review on the subject found that in 60% of the 170 studies considered, resistance to insecticides in insects targeted by these substances involved a cost in selective value, or fitness (Freeman *et al.*, 2021). It is likely that this type of result can be extrapolated to other species, including species not targeted by PPPs. However, this review highlights differences depending on the PPP considered. The cost of genetic adaptation to PPPs may also be reflected in greater vulnerability to parasites, as in *Daphnia* chronically exposed to the insecticide carbaryl (Jansen *et al.*, 2011), or to predators, as in *Culex pipiens* exposed to chlorpyrifos (Delnat *et al.*, 2019).

This question of the cost of adaptation to PPPs also arises when such adaptation is expressed at the community level. This is particularly the case for PPP tolerance induced at this scale by chronic exposure to these substances, mainly studied on microbial communities within the framework of the PICT approach. Indeed, the development of tolerance observed in these communities is generally accompanied by a decrease in microbial biodiversity and the inhibition of certain biological processes involving the activity of microorganisms. A physiological cost of tolerance to diuron acquired by aquatic phototrophic microbial communities chronically exposed to this herbicide has been demonstrated by recent work based on metabolomic analyses associated with physiological measurements (Lips *et al.*, 2022). All of these results highlight the need to develop more extensive knowledge and tools to better understand adaptation to PPPs and their associated costs at different biological scales (from the individual to the community, or even the ecosystem).

Furthermore, it would also seem relevant to address the issue of vulnerability to PPPs from the functional perspective. In this respect, various concepts, such as functional traits and their diversity, resilience, or functional redundancy, are important to consider, as highlighted by EFSA (EFSA Scientific Committee, 2016).

### Highlighting the different types of effects

The direct effects of PPPs on biodiversity (intraspecific, interspecific and functional) depend primarily on the toxic properties of the substances concerned, the use, fate and bioavailability of these substances in the environment, and the resulting exposure of organisms (see Chapter 2). In addition, these effects depend on the distribution of the degree of sensitivity of the different species within the exposed communities and their adaptive capacities (acclimatisation, tolerance, resistance, resilience, recovery), which concern various biological (populations, communities) and temporal scales (rapid and reversible physiological adaptation through developmental and phenotypic plasticity vs. longer term adaptation through selective evolutionary processes). Toxic effects can also have indirect consequences on biodiversity, by modifying the habitat and the interactions between species within and between trophic levels. All direct and indirect effects of PPPs on biodiversity influence ecological processes. The consequences of these effects on ecosystem functions depend mainly on the functional role of the impacted species.

The concepts outlined in the previous paragraph are commonly used to characterise the relationship between the toxicity of a substance and the observed variation in biodiversity, and are elaborated in this section. In addition to the direct or indirect nature of the effects, their degree of specificity with respect to one factor (e.g. PPPs) among a set of factors (e.g. habitat degradation and climate change) is also often discussed. Furthermore, as PPPs are designed to produce desired effects on organisms considered harmful, the distinction is also often made between targeted and non-targeted, or unintended/unwanted effects. Finally, as biodiversity is a dynamic entity, not every observed variation can be

automatically considered as an effect. Thresholds must therefore be defined to characterise the degree of variation that will be considered as an effect.

## ■ Direct and indirect effects

Direct effects are generally measured in terms of the consequences of the interaction of a PPP with biological processes on the physiological state and behaviour of individuals, and on the abundance and dynamics of exposed populations. Indirect effects refer to the impact of direct effects on certain species on other species that are either not exposed to or are resistant or less sensitive to the particular PPP. Since the end of the 20th century, the importance of the indirect impacts of PPPs has been increasingly highlighted in the literature, despite the difficulties of observing them in an experimental setting.

The existence of indirect effects can be inferred from field observations, based on correlations. Recent work has shown, based on data collected in European rivers, negative correlations between the toxicity of various identified contaminants (including PPPs) on photosynthetic organisms and the diversity of invertebrates living in these environments (De Castro-Catala *et al.*, 2020). As a further example, the direct effect of herbicide application on crops is to reduce a plant resource (weeds), and the indirect effect is a reduction in soil arthropods, with repercussions on bird populations, as these arthropods form the dietary basis of pheasant and grey partridge chicks (Taylor *et al.*, 2006). Beyond these correlations, the indirect effects of PPPs on biotic interactions (e.g. prey/predator relationships, competition) are mainly documented to date by experimental approaches.

Literature reviews on the impact of PPPs on wildlife emphasise the importance of indirect effects such as changes in food availability and/or interspecific competition for food (Boatman *et al.*, 2004; Kohler and Triebkorn, 2013). Other major indirect effects mentioned, which also concern biotic interactions and community effects, include host-parasite interactions, mainly described in amphibians. For terrestrial organisms, Gibbons *et al.* (2015) showed that indirect effects of PPPs are most often exerted in one of three ways: through the reduction of seed food for granivores following herbicide application; through the reduction of arthropod prey for insectivores following the application of insecticides or fungicides with insecticidal properties; or through the loss of insect host plants following herbicide application. This final point relates to the concept of habitat loss.

Finally, it is important to note that indirect effects are often the unintended consequences of a desired effect (e.g. food deprivation for beneficial organisms due to the elimination of competing weeds). They vary according to the intensity of the desired effect, its extent, duration and frequency of repetition, as well as the role in the ecosystem of the affected organisms. Thus, in the above example, the indirect effects on birds via arthropods that are victims of weed control are primarily dependent on the degree of weed control. They are therefore specific to the type of pressure (e.g. elimination vs. regulation) exerted on the target organisms, rather than to a particular substance.

## I Specific effects of PPPs

The issue of specificity of effects is raised, usually from a management perspective, to help identify responsibilities and measures to avoid unacceptable effects. The specificity of effects of PPPs refers to the portion of the observed effects attributable to the use of PPPs rather than to another factor. However, the factors that lead to the weakening of an ecosystem are not necessarily additive, but interact with each other. The impacts observed at complex levels of biological organisation are therefore most often linked to combinations of factors.

The specificity of effects is thus different from the selectivity of the mode of action of a substance, which is the ability of the substance to act on a narrow target without affecting other organisms. Otherwise, it is referred to as a broad-spectrum substance. This specificity of the mode of action is often based on the fact that the biological process affected is specific to the target organisms (e.g. blocking the production of an enzyme that is involved in vital processes for the target species, but which does not exist in others). This specificity, generally assessed on a theoretical basis at the time of marketing authorisation, is however often contradicted by subsequent observations, either because many physiological processes and mechanisms are common to living organisms, or because the targeted and tested effect is accompanied by other direct effects that were not initially suspected and not tested during the assessment (e.g. toxicity to birds and soil organisms from neonicotinoids targeting the nervous system of certain insects, and endocrine disrupting effects from organochlorines targeting nerve signal transmission), plus the indirect effects mentioned above.

## I Targeted, non-targeted or unintended effects

The concept of target organism or target effect is not a characteristic of the organism or effect under consideration, but depends on the intention of the user. Thus, the distinction may not be species-specific but rather location-specific. For example, a weed that is considered a pest within the cultivated plot may be a non-target species elsewhere (e.g. at the edge). The targeted effect is also sometimes referred to as the 'mode of action' of the substance, but the substance may have other effects that are not initially intended and/or not known.

The concept of non-target or non-intentional effects covers both direct effects (targeted effect or other effect on non-target organisms) and indirect effects (non-intentional effects on populations and communities impacted by direct targeted or non-target effects). Non-target effects are thus highlighted, in particular for species similar to target organisms (e.g. non-target insects for insecticides, non-target plants or algae for herbicides). However, they are also increasingly being demonstrated for more distant taxa, either through direct exposure, ingestion of contaminated prey, or through transformations of the food resource and/or habitat.

## I Effect thresholds

At the individual and population levels, effects are most often demonstrated by 'dose-response' studies in the laboratory. Model organisms are exposed to increasing doses or concentrations of PPP for a given time in order to characterise the response based on a

quantitative parameter (e.g. mortality). The expression of the relationship between exposure and effect is expressed by values such as the lethal concentration killing 50% of the batch of exposed individuals (LC<sub>50</sub>), the effective concentration reducing a given biological property by 50% (EC<sub>50</sub>; e.g. mobility or growth rate of a population of microorganisms) or the highest concentration tested for which no effect was observed in a chronic toxicity test (no observed effect concentration, or NOEC). Certain ratios can be calculated, such as the toxicity/exposure ratio (TER), the risk quotient (RQ) or the toxic units (TU), most often based on the EC<sub>50</sub> determined for the freshwater crustacean *Daphnia magna*. It is thus possible to determine threshold values to help interpret the results and highlight an effect or no effect. However, the relevance of these results depends on the choices made in the design of the study: species tested (to what extent the results are representative, and whether the effects observed in one species are transposed to another), exposure methods (a single substance or a mixture of substances, single or repeated exposure, one-off or chronic exposure, etc.), and the type of effect observed (e.g. physiological state indicator, population dynamics, the time frame over which it occurs, and whether or not it is reversible).

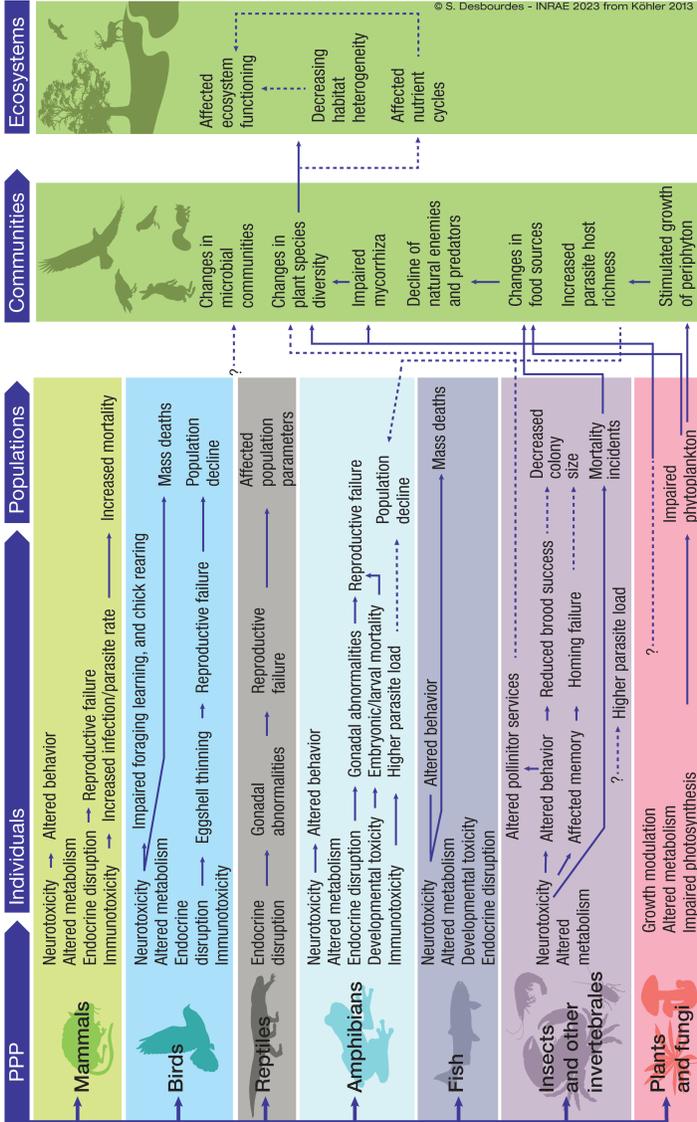
At the community level, the predicted or observed concentration is compared with standards such as the EQS (environmental quality standards of the WFD), the HC<sub>5</sub> (hazardous concentration for 5% of species), the ERL (environmental risk limit). The diversity of a community can be taken into account, in particular with the help of the species sensitivity distribution (SSD), which allows the calculation of potentially affected fractions (PAF) within a species community. These toxicity indices are a fundamental basis for the identification of effects. However, additional observations are necessary to take into account the exposure context (e.g. actual PPP application methods, landscape context, climate) and the representativeness of the species, not in terms of physiology, but in terms of characteristics and ecological role grouped under the concept of biological and functional traits (e.g. mobility, diet) and guild (organisms with similar traits).

## Effects on the state of biodiversity and its change

The impacts of PPP use on biodiversity are now widely recognised in Europe (Geiger *et al.*, 2010; Bruhl and Zaller, 2019) and increasingly around the world (Sanchez-Bayo and Wyckhuys, 2019; Sanchez-Bayo, 2021). Effects on biodiversity are generally analysed by group of organisms, level of organisation and environment. Figure 11, developed by Köhler and Triebkorn (2013), is an example of an illustration of the available knowledge of the dynamics of the spread of effects over a wide range of species at different levels of organisation, considering both direct effects and their indirect consequences on population kinetics. The many remaining unknowns are also noted.

Within the framework of the CSA, the bibliographic corpus analysed shows that, depending on the type of substance and the exposure conditions, effects are observed in almost all biological groups studied. However, it is more difficult to assess the resulting consequences in the various environments on population dynamics and community structure

Figure 11. Effects of PPPs at different levels of biological organisation (adapted from Köhler & Triebckorn, 2013)



The solid arrows represent known effects supported by evidence, and the dotted arrows represent plausible interactions between effects. Research is still needed where plausible interactions are not documented.

and thus, more generally, on biodiversity. In order to identify the main lessons drawn from this study, a distinction is made here whereby, in the first section, the biological groups for which the results show a major involvement of PPPs in the decline of populations are grouped together. The second part brings together the results suggesting a link between the use of PPPs and the population dynamics observed.

**PPP**s are a major contributor to the decline of certain taxonomic groups

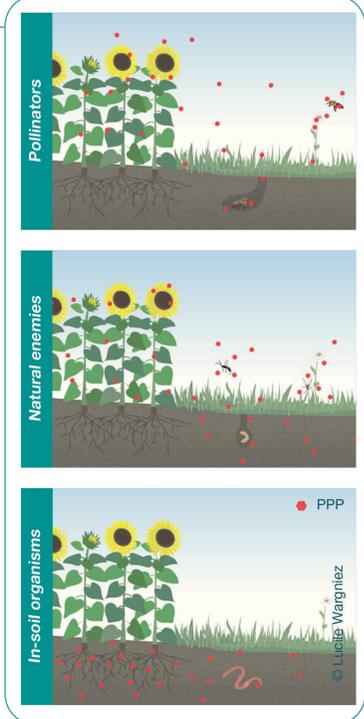
Terrestrial and aquatic invertebrates

The invertebrate group is a particularly diverse and abundant set of species, whose evolutionary dynamics are particularly difficult to document. As a result, they are largely under-represented in the work carried out by the IUCN on threatened species (invertebrate species with IUCN status represent only 2% of the species described, compared with 67% for vertebrates). However, the massive decline in the abundance of invertebrates over the last 50 years is recognised worldwide. For terrestrial invertebrates, the most recent scientific literature show a decline for 44% of species in Europe (Sanchez-Bayo and Wyckhuys, 2019). In freshwater environments in mainland France, IUCN data shows, for example, that 28% of freshwater crustacean species and 22% of mayfly species are threatened with extinction. In the case of terrestrial and freshwater molluscs, 11% of species are threatened.

The impact of PPPs has been particularly documented for terrestrial invertebrates. For example, according to Sanchez-Bayo and Wyckhuys (2019), chemical pollution, including PPPs, is the second most important cause of insect population decline after habitat loss due to urbanisation and intensive agriculture (and in some cases resulting from PPP use). The decline in terrestrial invertebrate diversity linked to the use of PPPs (Figure 12) is mainly observed in agricultural areas, where effects are observed in terms of abundance and species richness.

In terrestrial ecosystems, all taxa are affected, but lepidopterans (butterflies), hymenopterans (e.g. bees, bumblebees, ants) and coleopterans (e.g. ladybirds, carabid beetles) are the most affected (Sanchez-Bayo and Wyckhuys, 2019). Among PPPs, insecticides are the main ones responsible for direct effects on these organisms. One of the

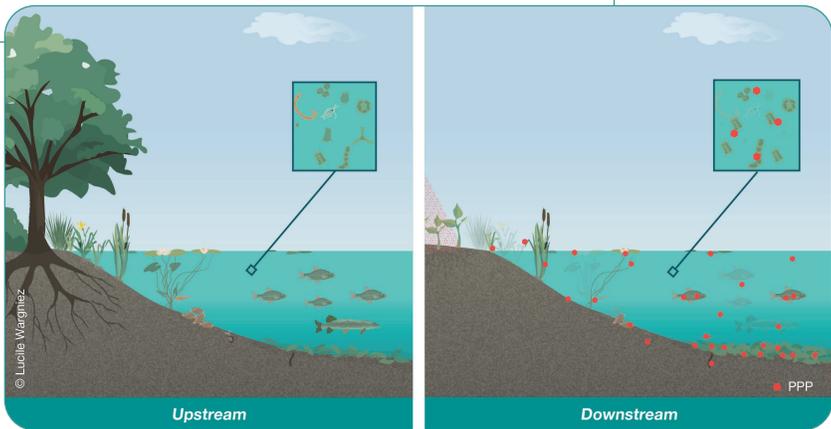
Figure 12. Exposure pathways of terrestrial invertebrates to PPPs (represented by red dots) in agricultural areas



consequences of the massive use of insecticides is a decrease in the abundance of natural enemies of crop pests. Their abundance is thus lower in conventional farming areas, and to a lesser extent in integrated farming, compared to what is observed in organic farming areas. In addition to the direct effects, there are indirect effects, mainly due to the impacts of herbicides on plant diversity and biomass and their consequences on habitats and trophic resources of terrestrial invertebrates (see figure 19, p. 109 for illustration).

In agricultural environments, marked effects of PPPs on the biodiversity of macroinvertebrates in waterways have also been observed, as illustrated in Figure 13. They result in the disappearance of some species or a decrease in their abundance downstream from cultivated areas, with consequences, for example, on the decomposition of plant litter, which may then be strongly, or even completely, inhibited. Thus, on a European scale, it is estimated that PPP contamination leads to losses of up to 40% of the species richness (or number of species) of river macroinvertebrates (Beketov *et al.*, 2013).

**Figure 13. Presence of PPPs in rivers and their upstream and downstream effects**



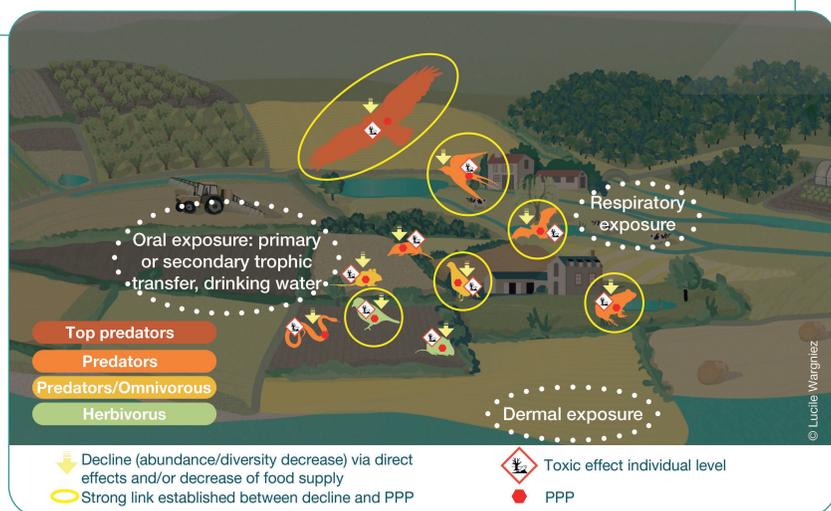
Impacts on the diversity of terrestrial and aquatic invertebrates have mainly been documented with regard to insecticides, with neonicotinoids and pyrethroids appearing to be the families of chemicals still in use that are of greatest concern. In particular, insecticides are blamed for the poor ecological quality of 30% of small European rivers, posing a significant risk to aquatic macroinvertebrates (Kattwinkel *et al.*, 2011). Herbicides also contribute to the decline of these macroinvertebrates, via indirect effects through their impact on habitats and food resources provided by macrophytes and other plant species.

### Birds and bats

The IUCN red list based on 2016 data indicates that of the 284 bird species that breed in France, 92 are threatened, which represents a deterioration of the situation since 2008 (increase in threatened species from 26% to 32%). This deterioration is also greater than

that observed on a global scale, where 12% of species are threatened<sup>17</sup>. PPPs are identified as one of the major factors in the decline of bird abundance and diversity in agricultural areas, interacting with landscape homogenisation and intensification of farming systems. The indicator of relative abundance of bird populations according to their environmental preference, calculated by the *Suivi temporel des oiseaux communs* (Temporal Monitoring of Common Birds) programme (STOC), shows a 29.5% decline in this indicator in agricultural areas between 1990 and 2020, compared with 27.6% in built-up areas, 9.7% in forest areas, and a 19.4% increase for generalist birds<sup>18</sup>. Depending on the bird species and their diet, the impact of PPPs results primarily either from a direct effect (e.g. ingestion by granivorous birds of treated seeds or consumption of PPP-contaminated prey) or from an indirect effect (e.g. reduction in food resources following a decline in prey; Figure 14).

**Figure 14. Effects of PPPs on terrestrial vertebrates in agricultural areas (adapted from Pesce *et al.*, 2023a)**



Phytopharmacovigilance networks in various European countries (including, for example, France, England and Spain) reveal a large number of cases of PPP poisoning of birds in the vicinity of agrosystems. Since the early 2000s, granivorous birds have constituted the majority of cases of direct poisoning, following the ingestion of seeds coated with neonicotinoid insecticides (especially imidacloprid), and more rarely with other substances such as fungicides. Various European crops are subject to the application of these substances

17. <https://uicn.fr/wp-content/uploads/2016/09/Liste-rouge-Oiseaux-de-France-metropolitaine.pdf> (accessed 9/01/2023).

18. [https://www.vigienature.fr/sites/vigienature/files/atoms/files/syntheseoiseauxcommuns2020\\_final.pdf](https://www.vigienature.fr/sites/vigienature/files/atoms/files/syntheseoiseauxcommuns2020_final.pdf) (accessed 9/01/2023).

(wheat, barley, corn, rapeseed, sunflower, peas, flax, and soybeans), leading to potential exposure of a large number of species, but autumn sowing has been identified as leading to a higher risk of neonicotinoid-induced mortality. Possible explanations include the fact that the majority of omnivorous birds focus their diet on seeds at this time. Exposure through trophic pathways in insectivorous birds was recently demonstrated for swifts in Switzerland on the basis of multi-residue analyses of food boluses for nestlings, suggesting the possible existence of impacts related to the consumption of contaminated prey (Humann-Guilleminot *et al.*, 2021).

For insectivorous birds, the impact of PPPs is mainly expressed indirectly, through the decline in food resources. Several European studies have demonstrated a relationship between the use of PPPs and the concurrent decline in insect communities and bird populations. For example, by reducing the arthropod food resource, insecticides applied during the breeding season have been identified as one of the factors reducing the reproductive performance of corn buntings (*Miliaria calandra*) and yellowhammers (*Emberiza citrinella*) (Brickle *et al.*, 2000; Boatman *et al.*, 2004; Hart *et al.*, 2006). Regarding more specific chemical families, a review of the literature highlighted the likely important role of indirect effects from neonicotinoids and fipronil via a reduction in the food supply for birds in field crops (Gibbons *et al.*, 2015).

Neonicotinoids are therefore especially implicated in the decline of granivorous and insectivorous birds. These observations are supported by scientific evidence of negative correlations between the abundance of several breeding bird species and the use of neonicotinoids (Lennon *et al.*, 2019) or the concentration levels of these substances in surface waters (Hallmann *et al.*, 2014). While controlling for other factors associated with agricultural intensification (land use change, area cultivated, fertilisers), the significant influence of imidacloprid concentrations on the decline of six out of fifteen bird species studied (Eurasian skylark, barn swallow, western yellow wagtail, common starling, common whitethroat, mistle thrush) has thus been demonstrated in the Netherlands (Hallmann *et al.*, 2014). Furthermore, disruption of flight efficiency and orientation has emerged as a significant outcome of exposure and sublethal effects of neonicotinoids in migratory birds. These sublethal effects could lead to the alteration of the migration success of birds using agricultural areas as staging areas.

As regards chiropterans (bats), the literature more generally suggests a negative impact of organophosphate/carbamate PPPs (such as chlorpyrifos) and organochlorine PPPs (DDT and lindane), which have now been banned, and of pyrethroids (used both in agriculture and for wood treatment) on the population dynamics and diversity of bats, which have undergone severe declines since the middle of the 20th century. Of the 34 bat species present in France, 8 are now classified as threatened by the IUCN and 8 others as near-threatened<sup>19</sup>. PPPs have been identified as one of the potential causes of this decline, but knowledge is currently too incomplete to characterise the population impacts of the

19. <https://uicn.fr/wp-content/uploads/2017/11/liste-rouge-mammiferes-de-france-metropolitaine.pdf> (accessed 9/01/2023).

substances currently used. However, as for birds, altered movements have been observed in one species of Asian bat (*Hipposideros terasensis*) following repeated exposure to imidacloprid. To our knowledge, no data published to date confirm the existence of such effects on chiropteran populations in the field, but a recent study supports this behavioural data and suggests that the alteration of their echolocation orientation system following exposure to this type of insecticide probably affects their movements and hunting activities (Wu *et al.*, 2020).

For raptors, mortality linked to deliberate and illegal poisoning by anticholinesterase insecticides (applied to meat baits) and to the consumption of contaminated prey following the application of anticoagulant rodenticides is particularly well documented. Reported throughout the world, these cases of poisoning are associated with mortalities that may involve from a few individuals to several hundred, the species affected most often being scavenging birds of prey. In France, the consequences of deliberate poisoning with anticholinesterase drugs have been reported for several species of vultures and for the red kite (Beryn and Gaillet, 2008; Beryn *et al.*, 2015). For the latter, in similar contexts to France, population declines linked to these illegal practices have been estimated in Spain to be between 20 and 40% (Mateo-Tomas *et al.*, 2020). It is difficult to define a trend in the occurrence of these practices since the beginning of the 21st century, but several articles documenting recent cases have been published in recent years, showing that these practices remain an ongoing problem on a national and global scale. In different contexts, the agricultural use of anticoagulant rodenticides has also led to secondary and lethal poisoning of raptors in different parts of the world, including mainland France (Beryn and Gaillet, 2008; Coeurdassier *et al.*, 2012; 2014a; 2014b) and Reunion Island (Coeurdassier *et al.*, 2019). While these examples clearly demonstrate that raptors are vulnerable to PPPs, knowledge about the levels of exposure of these birds to currently used substances and the resulting effects on their populations is to date almost non-existent.

## Amphibians

Amphibians are among the taxonomic groups most affected by the current massive reduction in biodiversity at the global level (Ockleford *et al.*, 2018). According to the IUCN red list for France, 8 out of 35 amphibian species are threatened in metropolitan France, and populations are declining for 60% of amphibians<sup>20</sup>. Various factors have been identified as contributing to these declines, including habitat destruction, changing conditions related to climate change, pathogens and the introduction of invasive species, but also PPPs. Declines in amphibian populations have notably been linked to high prevalences of diseases, some of which could be favoured through exposure to PPPs due to direct toxic effects of an immunotoxic or endocrine disrupting kind, as well as indirect effects via changes in the dynamics of pathogens or parasites and their various vectors and hosts. Episodes of

20. [https://uicn.fr/wp-content/uploads/2015/09/Liste\\_rouge\\_France\\_Reptiles\\_et\\_Amphibiens\\_de\\_metropole.pdf](https://uicn.fr/wp-content/uploads/2015/09/Liste_rouge_France_Reptiles_et_Amphibiens_de_metropole.pdf) (accessed 9/01/2023).

mortality, developmental problems and reproductive failure following exposure to PPPs, even at low doses and for substances currently in use, have also been suggested.

However, in their review on this topic, Mann *et al.* (2009) describe the difficulties (which still remain) in establishing clear links between amphibian population declines and the toxic effects of PPPs, due in particular to the many confounding factors (e.g. climatic factors, constantly changing environments, changes in land use and practices) which make it difficult to establish robust cause-effect links. Moreover, in the case of amphibians, this task is made more complex by the fact that exposure involves both the aquatic and terrestrial environment, and that it is very difficult to quantify the proportion of PPP effects linked to strictly aquatic, strictly terrestrial, or mixed exposure (Ockleford *et al.*, 2018).

## ■ The impacts of PPPs on other taxonomic groups

### Primary producers and heterotrophic microorganisms

Knowledge of the effects of PPP contamination of terrestrial and aquatic environments on the biodiversity of primary producers and heterotrophic microorganisms is rather fragmentary and is based mainly on experimental studies under controlled conditions, which makes generalisation difficult. However, some observations can be made on the basis of the analysis of the literature.

Because of their phytotoxic modes of action, herbicides, and in particular photosynthesis inhibitors, most of which are no longer used in France (and more generally in Europe), appear to be the most harmful substances for the biomass and diversity of higher plants, but also for lichens, microalgae and cyanobacteria on land and in water. Residual substances in the marine environment also contribute to the degradation of coral reefs and mangroves through direct and indirect effects. It should be noted, however, that there is a lack of knowledge about the effects of PPPs on the diversity of marine phototrophic organisms in comparison with those of freshwater.

Effects of non-photosynthesis inhibiting herbicides on higher plants have been demonstrated at concentrations well below the approved field dose. Because of the impact of herbicides on non-target plant communities, the management of invasive plant species by this type of PPP is now strongly questioned, as it may lead to the weakening or elimination of some local endemic species, thus counterproductively favouring the targeted invasive species.

Concerning phototrophic microbial communities, the impact of herbicides on their structure and diversity has notably been demonstrated *in situ* in various contaminated terrestrial and aquatic ecosystems, using the PICT method. This approach made it possible to highlight, firstly, the replacement within natural microbial communities of species that are the most sensitive to certain herbicides (particularly from the triazine and phenylurea families) by species that are more tolerant to these substances, and secondly, a return of these sensitive species when the toxic pressure decreases. This loss of tolerance can be facilitated by the downstream migration of sensitive species from less exposed upstream

areas, thus underlining the importance of maintaining refuge areas and connectivity between habitats as mentioned above (see 'Remediation' section).

In addition to herbicides, copper is also a substance of concern, particularly with regard to phototrophic and heterotrophic microorganism biodiversity, which can be affected by chronic exposures at concentrations in the order of those found in the environment. The PICT approach has also been successfully used to demonstrate the effects of copper on these microorganisms in soil and aquatic environments, including the sediment matrix. In general, fungicides (particularly sterol biosynthesis inhibitors, strobilurins and copper) have the greatest effect on heterotrophic microbial communities in soils and aquatic environments. *In situ*, the strongest effects have been observed on aquatic fungal communities (hyphomycetes) involved in the degradation of plant litter.

### Terrestrial and aquatic vertebrates

While many studies have documented the exposure of terrestrial and aquatic vertebrates to PPPs and their contamination by a wide range of substances, the resulting effects at the population level (and in some cases at the individual level) remain largely unknown (except for birds and, to a lesser extent, bats and amphibians; see section on 'Specific effects of PPPs'). The role of PPPs in the decline of reptiles is suspected (Ockleford *et al.*, 2018), but to date the scientific evidence is too limited to establish this.

For other vertebrates, the lack of knowledge makes it impossible to determine the effects of PPPs at the population level. This can be explained either by the difficulty of conducting studies (e.g. marine mammals and terrestrial megafauna), or by the fact that the vast majority of ecotoxicological studies on the effects of PPPs are based on exposures carried out under controlled conditions associated with response measurements at the individual and sub-organism levels. This makes it difficult to transpose the results to the population level in contaminated ecosystems. Thus, while it is possible to conclude that some of these substances can induce effects on species exposed under experimental conditions, their potential to affect individuals and populations in the natural environment generally remains to be demonstrated.

## Impacts on ecosystem functions

The bibliographical review has shown that the effects of PPPs on ecosystem functions are generally approached through the biological activities and ecological processes in which the impacted organisms are involved. Highlighting the resulting consequences on the functioning of ecosystems implies considering the relationships between the state of biodiversity and its functions. In particular, it is a question of integrating the relative importance of the different processes that support these functions, and also the fact that certain constituent states of ecosystems that determine their functionality do not depend on biodiversity. Ecosystem functions are more rarely chosen as an object of analysis per se, and if so, are approached with concepts and vocabulary that may be specific to the types of organisms

and environments considered. In order to assess the body of knowledge that covers these diverse aspects, a classification of these functions was established within the framework of the CSA (Table 2), in relation to the ecological processes that are used to assess the impacts of PPPs on ecosystems, and based on classifications previously proposed in the literature.

On the basis of this classification, the results documenting the impacts of PPPs on ecosystem functions are summarised in the following sections.

### ■ Relationship between biodiversity effects and ecosystem functions

Some ecosystem characteristics that determine their functionality do not depend on biodiversity, such as geological and climatic characteristics. However, increases in species richness and population abundance are generally accompanied by increases in functional diversity. The effects of PPPs on organisms and communities thus have implications for the ecological processes in which they are involved, and for the ecosystem functions that these processes support.

Figure 15 illustrates the links between PPP uses, biodiversity status, ecological processes and ecosystem functions.

This figure shows the importance of addressing the functional role of species impacted by the effects of PPPs, the degree of functional redundancy, i.e. the degree of functional redundancy, i.e. the substitutability between impacted and non-impacted species to fulfil the same function, and interactions between species. In addition, some PPPs are designed to specifically target biological groups that directly contribute to certain ecological processes, for example photosystem inhibiting herbicides (such as triazines and phenylureas) that directly affect photosynthesis and primary production (Black, 2018). These targeted functional effects can strongly influence the relationships between biodiversity and ecosystem functioning through feedback mechanisms from ecological processes and ecosystem functions to biodiversity. These feedbacks are poorly studied and remain relatively unknown (Duncan *et al.*, 2015; Grace *et al.*, 2016; Qiu *et al.*, 2018; van der Plas, 2019).

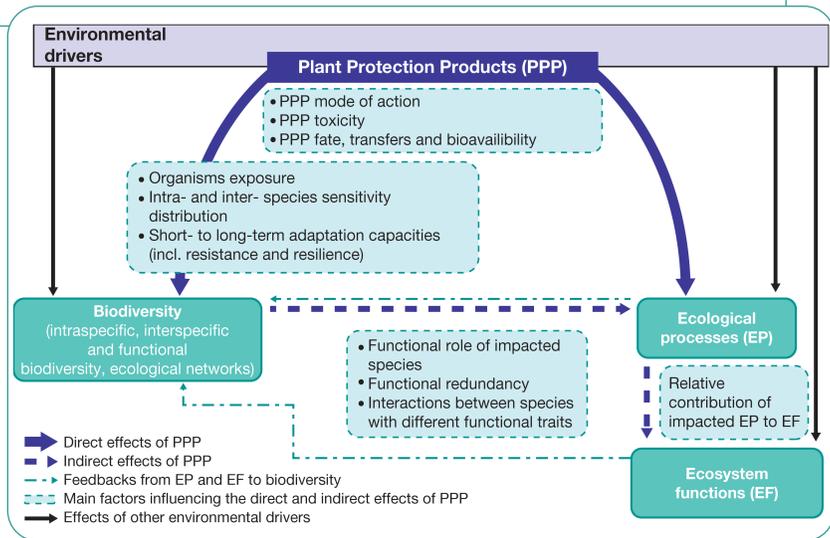
Moreover, the relationships identified between biodiversity and functions are rarely linear in nature. While it can be found that the abundance of a functional group is favourable to the dynamics of an ecological function (e.g. the more pollinators there are, the more pollination increases), this link also depends on equilibria relative to the context (beyond a threshold, the overabundance of pollinators in relation to the floral resource will no longer improve pollination). Functions are based on equilibria, optima and complementarities, rather than on linearly positive or negative relationships with the abundance of a given species or group. Thus, species richness is not sufficient to guarantee the functional resilience of an ecosystem, in the event that certain functions are only provided by species that are negatively impacted by the pressure exerted.

**Table 2. Proposed CSA classification of ecosystem functions potentially affected by PPPs (adapted from Pesce *et al.*, 2023b)**

	Ecosystem functions	Definitions	Examples of functional parameters used in ecotoxicology
F1	Regulation of gas exchange	Production and consumption of gases and regulation of gas exchange between different environmental compartments	Photosynthesis, respiration, methanogenesis, denitrification, nitrogen fixation, evapotranspiration
F2	Dissipation of contaminants in terrestrial and aquatic ecosystems	Filtration, buffering, sequestration and degradation of chemical and biological contaminants and wastes	Biodegradation and phytodegradation potential, enzymatic activity potential, exopolysaccharide production
F3	Resistance to disturbance	Mitigation of and resilience to environmental (heat waves, fires, storms, floods, mudslides, avalanches) and anthropogenic (pollution) disturbances	Biomass of above-ground (cover) and below-ground (root systems) terrestrial vegetation, biomass of aquatic biological structures (e.g. coral reefs, seagrass beds, mangrove vegetation), pigment production, exopolysaccharide and mucilage production
F4	Water retention in soils and sediments	Water retention and storage in soil and sediments to preserve freshwater resources	Bioturbation of soils and sediments, exopolysaccharide and mucilage production, root architecture
F5	Regulation of water flows	Regulation of water runoff and discharge	Bioturbation of soils and sediments, exopolysaccharide and mucilage production, root architecture
F6	Albedo	Regulation of surface reflectivity by vegetation	Biomass and plant cover, macroalgal and phytoplankton biomass, pigment production
F7	Production and supply of organic matter in terrestrial and aquatic ecosystems	Production and dispersal of biomass and organic matter that can serve as energy sources in food webs	Primary production, secondary production
F8	Regulation of nutrient cycles in terrestrial and aquatic ecosystems	Decomposition of organic matter; transport, storage and recycling of nutrients	Methanogenesis, nitrification, denitrification, enzymatic activity, decomposition of particulate organic matter
F9	Formation and maintenance of soil and sediment structure	Role of biota in the formation and maintenance of soil and sediment structure (including shorelines and coasts)	Bioturbation of soils and sediments, biomass of above-ground (cover) and below-ground (root systems and mucilage) terrestrial vegetation, aquatic biomass (e.g. coral reefs, seagrass beds, mangrove vegetation), production of microbial filaments and exopolysaccharides
F10	Dispersal of propagules in terrestrial and aquatic ecosystems	Role of biota in the release and movement of propagules (including floral gametes and seeds, aquatic and marine spores, eggs and larvae)	Sexual (e.g. pollination) and vegetative reproduction of plants, production of spores (including akinetes), transport of propagules by terrestrial and aquatic organisms

Ecosystem functions	Definitions	Examples of functional parameters used in ecotoxicology
F11 Provision and maintenance of biodiversity and biotic interactions in terrestrial and aquatic ecosystems	Provision and maintenance of biodiversity and interactions within biotic communities to maintain ecosystem functioning, contain the impact of epidemics or harmful outbreaks (e.g. by controlling populations of potential pests and disease vectors), ensure the production and use of natural materials (biological and genetic resources) that can be used by organisms for their health, and contribute to a self-sustaining diversity of organisms developed through evolution (and capable of continuing to change)	Population and community dynamics, trophic interactions, competition, facilitation, parasitism, symbiosis, genetic potential, production of nutrients, hormones and biocides
F12 Provision and maintenance of habitats and biotopes in terrestrial and aquatic ecosystems	Provision of appropriate living spaces for natural biological communities and individual species, including for reproduction and rearing, as well as refuges and corridors within and between natural and semi-natural ecosystems (connectivity)	Bioturbation of soils and sediments, biomass and diversity of above-ground (cover) and below-ground (root systems and mucilage) terrestrial vegetation, terrestrial and aquatic biogenic structures

**Figure 15. Expected impacts of PPPs on biodiversity, ecological processes and ecosystem functions through their interrelationships (adapted from Pesce *et al.*, 2023b)**



## I Proven impacts on ecosystem functions

Based on knowledge from the literature, it is possible to identify impacts of different PPPs on most of the categories of ecosystem functions defined in Table 2, both in terrestrial and aquatic environments. However, certain categories of functions are more frequently studied, such as regulation of gas exchange (F<sub>1</sub>), dissipation of contaminants (F<sub>2</sub>), resistance to disturbance (F<sub>3</sub>), production of organic matter (F<sub>7</sub>), regulation of nutrient cycles (F<sub>8</sub>), dispersal of propagules (F<sub>10</sub>), provision and maintenance of biodiversity and biotic interactions (F<sub>11</sub>) and provision and maintenance of habitats and biotopes (F<sub>12</sub>).

### Effects of PPPs on the regulation of gas exchange

The effects of PPPs on the regulation of gas exchange (F<sub>1</sub>) are mainly addressed through the study of three ecological processes, photosynthesis, atmospheric nitrogen fixation and aerobic respiration, via measurements of potential or actual activity. These include a) photosynthetic activity of the various primary producers (including microorganisms) contributing to CO<sub>2</sub> consumption, b) nitrogenase activity in bacteria, cyanobacteria and plants, which allows the fixation of atmospheric nitrogen N<sub>2</sub>, and c) O<sub>2</sub> consumption and/or CO<sub>2</sub> production (measured mainly in heterotrophic microorganisms) in the context of aerobic respiration.

Herbicides are clearly the organic PPPs with the strongest impact on the photosynthetic activity of all primary producers and on the nitrogenase activity of plants and cyanobacteria. However, the herbicides for which the strongest effects have been demonstrated, in particular the photosystem II inhibitors (e.g. triazines, substituted ureas), are now banned from plant protection use, even though they are still present in the environment (see section 'Transfer dynamics and fate of substances'). Furthermore, it has been shown that environmental concentrations of copper can have a negative impact on the regulation of gas exchange by phototrophic and heterotrophic microbial communities in terrestrial and aquatic environments. For instance, copper has been found to affect the photosynthetic activity of microalgae and cyanobacteria in continental and marine aquatic environments, as well as the nitrogenase activity of cyanobacteria and microbial respiration in soils and aquatic environments, including sediment. However, the effects of copper are highly variable depending on the properties of the habitat and the level of sensitivity of the microbial communities, which appear to have a high capacity to adapt to this toxicant, thus increasing their level of tolerance and resilience.

### Effects of PPPs on the dissipation of contaminants

With respect to the dissipation of contaminants (F<sub>2</sub>), it appears that the capacity of terrestrial and aquatic microbial communities to biodegrade certain PPPs can be increased by prolonged and/or repeated exposure to these substances. This increase reflects a functional adaptation that results from structural changes due to the selection of microorganisms initially possessing biodegradation capacities and/or horizontal transfers of genetic variants that promote this microbial process. This type of functional adaptation has been demonstrated with different substances such as herbicides of the phenylurea

family, triazines, and insecticides of the carbamate family. It has been shown, in the case of atrazine, that this degradation capacity could be maintained in certain bacterial populations for several years in the absence of treatment via the conservation of degradation genes (Yale *et al.*, 2017). However, to date, it has proved difficult to quantify the influence of this biodegradation activity on the attenuation of PPPs in different matrices in the field, or to assess its consequences on the contamination of environments by transformation products, the concentrations and ecotoxicological effects of which are poorly understood. Moreover, as mentioned previously (see section '14.2. Effects of accumulations and mixtures of substances'), there is the issue of the biological cost of such adaptation at the scale of the communities in which the populations capable of degrading PPPs develop. Moreover, this type of adaptation can be inhibited by the presence of other PPPs (e.g. impact of Bt toxins, Accinelli *et al.*, 2004, or copper, Dousset *et al.*, 2007, on the degradation of glyphosate) and is influenced by various environmental factors such as temperature or the presence of exogenous organic matter.

### Effects of PPPs on resistance to disturbance

The effects of PPPs on resistance to disturbances (F3) are relatively little addressed in the literature. However, studies examining disturbances associated with climate change (e.g. increased mean temperatures and temperature fluctuations, increased intensity of precipitation and drought, flooding, ocean acidification) show that these often increase the sensitivity of organisms (and the vulnerability of populations) to environmental toxins (including PPPs) and, in turn, these substances may reduce the ability of organisms to cope with the consequences of climate change. For example, the vast majority of studies that have combined temperature increase and PPP exposure have shown a synergistic interaction of these factors. Furthermore, sublethal effects of PPPs can lead to a reduction in locomotor ability, thus increasing vulnerability to predation (e.g. observed in various vertebrates following exposure to neurotoxic PPPs such as organophosphates and carbamates; Lambert *et al.*, 2005) or to climate change (e.g. decrease in the ability to expand northwards in the damselfly *Coenagrion scitulum* exposed to the pyrethroid insecticide esfenvalerate at the larval stage; Dinh *et al.*, 2016).

### Effects of PPPs on organic matter production

The production of organic matter (F7), is mainly the result of primary carbon production by photosynthetic organisms, and in particular plants and micro-organisms possessing chlorophyll a. The overall amount of carbon fixed is relatively similar in land (terrestrial and aquatic) and marine ecosystems. However, the biomass and turnover time of primary producers are very different between marine environments (faster turnover) and terrestrial environments (higher biomass). As expected, given the ecological role of primary producers, the PPPs that have the greatest impact on this type of organism through direct effects (e.g. herbicides, copper) are those that have the greatest impact on the production of organic matter. The impact of these PPPs on these organisms decreases their biomass and changes their nutritional quality, which may affect higher trophic levels.

## Effects of PPPs on the regulation of nutrient cycles

The regulation of nutrient cycles (F8) is strongly disrupted by PPPs, as many substances inhibit the degradation processes of organic matter in the different matrices that make up terrestrial and aquatic environments. The literature mainly reports the effects on the fragmentation of particulate organic matter (plant litter) by both microorganisms (fungal and bacterial communities) and invertebrates. Negative effects on the microbial decomposition of this particulate organic matter are mostly observed during the application of fungicides, in particular tebuconazole, azoxystrobin, chlorothalonil or copper. Rasmussen *et al* (2012), for example, found PPPs to inhibit the microbial decomposition of plant litter in streams located in agricultural catchment areas compared to those located in forest areas, despite a possible positive effect of nutrients from agricultural fertilisers on this decomposition process. The degradation activity of terrestrial and aquatic plant litter by invertebrate communities is also impacted by PPPs and other toxic substances. The specific impacts of PPPs (including herbicides, fungicides and insecticides) on the degradation activity of invertebrates could be illustrated in a study carried out on different watercourses in an agricultural context in south-western France subject to PPP pressure (Brosed *et al.*, 2016). In aquatic and terrestrial environments, among PPPs, insecticides and also copper are the main agents responsible for inhibiting this activity in invertebrates. For example, a study carried out in German rivers located in wine-growing areas showed that copper contamination, resulting from its fungicidal use, has a strong effect on litter degradation (up to 100% inhibition in the most contaminated sites), an effect potentially mitigated by a replacement of species within the detritivore crustacean communities (Fernandez *et al.*, 2015). In terrestrial environments, Pearsons and Tooker (2021) showed that seed coating with neonicotinoids or application of a pyrethroid insecticide significantly affected this ecological process, while Martinez *et al.* (2016) showed that sublethal concentrations of copper could have the same type of effect.

Furthermore, the effects of PPPs on the regulation of nutrient cycles, via the microbial decomposition of dissolved organic matter, are also being studied extensively in the soil and continental aquatic environments (surface water and sediment) through the measurement of various enzymatic activities involved in biogeochemical cycles (C, N, P, S). It is very difficult to draw conclusions on this subject, as the microbial responses observed are generally highly variable (from inhibition to stimulation) depending on the environmental context and the temporal scale studied (see for example the meta-analysis by Nguyen *et al.*, 2016, on the effects of glyphosate in soils). However, a certain consensus can be observed concerning the effects of copper, which inhibits a wide range of heterotrophic microbial activities involved in biogeochemical cycles, both in terrestrial and aquatic environments.

## Effects of PPPs on the dispersal of propagules

The dispersal of propagules (F10) is also affected by PPPs in terrestrial and aquatic ecosystems. The vast majority of our knowledge on this subject concerns pollination, particularly because of the proven role of PPPs in the decline of insect populations, including pollinating insects. However, it appears that relatively little research has assessed the effects

of PPPs on the pollination process itself, with the majority of the literature limited to the study of pollinators without actually measuring their pollination activity (Uhl and Bruehl, 2019). Despite this, it has been shown that exposure to PPPs, and neonicotinoid insecticides in particular, can reduce pollination in different agricultural contexts such as apple growing (Stanley *et al.*, 2015) or grape growing (Brittain *et al.*, 2010). Several studies conducted in different crop types also show that flower visitation rates and pollination intensity are higher in organic farming systems than in conventional systems. However, this still needs to be substantiated, as the impact of PPPs on pollination can be influenced by different factors such as the life history traits of pollinating insects and the degree of specialisation of plant-insect interactions. This impact may also be indirect (i.e. not related to direct toxicity to pollinating organisms), for example in response to the decline in plant diversity or inhibition of flowering of higher plants induced by herbicide exposure.

Furthermore, PPPs, and in particular fungicides, also have an impact on the sporulation of hyphomycetes in aquatic environments. Inhibition of this process has been demonstrated for various synthetic fungicides, including azoxystrobin and various nitrogenous substances such as tebuconazole and clotrimazole, but also for copper.

### Effects of PPPs on the provision and maintenance of biodiversity and biotic interactions

The effects of PPPs on the provision and maintenance of biodiversity in terrestrial and aquatic ecosystems (F11) are described in the section 'Effects on biodiversity status and trends'. Beyond these effects, the literature also shows an impact of PPPs on numerous biotic interactions in these ecosystems. These include interactions between plants and pollinators, but also between plants and symbiotic microorganisms that are associated with their roots and promote their growth, such as mycorrhizae and nitrogen-fixing rhizobacteria. Most studies on this subject only deal with the effects of PPPs on microorganisms, without considering the impacts on plants. Although some results are sometimes contradictory, the few studies that address the effects of PPPs on plants through the response to PPPs of symbiotic microorganisms associated with the roots generally show a negative impact of these substances, in particular fungicides through mycorrhizae, and insecticides and fungicides (including copper) through rhizobacteria. In the overseas territories studied, the contamination of coral reefs by several herbicides also raises questions, given that hermatypic corals are highly dependent on their symbiosis with zooxanthellae (dinoflagellates of the genus *Symbiodinium*). The latter are highly sensitive to these substances (in particular to photosystem II inhibitors such as substituted ureas and derivatives of triazines that were previously used and are still present in these environments), with partial inhibition of their photosynthetic activity at concentrations in seawater of less than  $\mu\text{g/l}$ . Various terrestrial and aquatic animals may also be affected by PPPs as a result of effects on their microbiota. Although this area of research is developing rapidly, these effects remain relatively unexplored. However, recent work has shown the influence of various PPPs on different types of microbiota. One example is the modification of the microbiota of honey bee larvae by the herbicide glyphosate (Motta *et al.*, 2020; Castelli *et al.*, 2021)

or by the fungicides carboxamide and boscalid in the context of infection by the parasite *Nosema ceranae* (Paris *et al.*, 2020). Cases of dysbiosis have also been described in birds, mammals or amphibians (adult stage) exposed to glyphosate or the insecticide trichlorfon. Two recent reviews provide a comprehensive and detailed overview of current knowledge, which is very much confined to laboratory model animals (Chiu *et al.*, 2020; Syromyatnikov *et al.*, 2020).

In addition, several studies suggest an increase in the vulnerability of certain populations to parasites or pathogens in response to exposure to various PPPs (e.g. neonicotinoid insecticides for wild bees and bats; organochlorine insecticides for various amphibian species; glyphosate herbicide for certain fish; various PPPs for Pacific oysters). Some authors have attributed the increase in bacterial diseases in birds over the last two decades to impaired immunity due to exposure to PPPs, in particular neonicotinoids. Sublethal effects of PPPs can also induce a reduction in locomotor ability, leading to increased vulnerability to predation (e.g. observed in various vertebrates following exposure to neurotoxic PPPs such as organophosphates and carbamates, Lambert *et al.*, 2005). As discussed above ('Effects of accumulations and mixtures of substances' section), the cost of genetic adaptation to PPPs may also result in increased vulnerability to pests.

Furthermore, several studies have looked at the impacts of PPPs on biotic interactions in relation to the natural regulation of crop pests, an ecosystem service. In many cases they have shown a negative effect on beneficial organisms and/or their ability to consume prey. The trophic cascade phenomenon has also been suggested in relation to rodenticides, with the risk of losing the biological regulation service provided by predators on the pests that are targeted by treatments. In this context, the use of PPPs to maintain low pest densities would decrease the populations of their natural predators, causing pest population dynamics to be predominantly controlled by PPP use due to a decrease in natural control. More generally, PPPs can modify the biotic interactions involved in bottom-up (resource-regulated) and top-down (consumer-regulated) relationships, which are known as 'vertical' relationships, and in competitive relationships, which are known as 'horizontal' relationships, although the latter are less studied. The type of vertical regulation (i.e. bottom-up or top-down) involved in the propagation of PPP effects varies according to the composition of food webs and the type of PPP considered (and the sensitivity of organisms at different trophic levels). Seasonal variations can thus be observed, depending on the life cycles of the organisms and the timing of the use of the different PPPs.

### Effects of PPPs on the provision and maintenance of habitats and biotopes

In addition to above-mentioned biotic interactions, the impact of PPPs on certain organisms can result in changes to habitats and biotopes (F12). In particular, the impacts of herbicides on the diversity and biomass of plants not targeted by the treatments (especially during the flowering period) can reduce the habitats of terrestrial invertebrates.

For example, this is particularly well documented for glyphosate. Indirect adverse effects of this herbicide on various invertebrates have been described due to changes in their habitat as a result of direct effects on vegetation. The latter includes, in particular, specific host plants necessary for the reproduction and development of butterflies, the canopy that shelters various dependent invertebrates (e.g.: araneids, beetles, diptera) and vegetation suitable for the presence and survival of various spiders. This observation also applies to aquatic environments, where studies have revealed herbicides indirectly affecting invertebrates due to biotope and habitat modification resulting from a direct effect on macrophytes. In the marine environment, de Caralt et al (2020) suggest that the decline of fucoid meadows (brown macroalgae) observed in the Mediterranean may be partly attributable to the contamination of the coastal zone by PPPs (in particular herbicides and copper). These meadows play a major ecological role in shallow benthic ecosystems on rocky Mediterranean coasts, particularly in terms of habitat structuring.

### Innovations and future directions for the assessment of effects

The progressive increase in the number of studies carried out in natural environments (e.g. epidemiological and ecotoxicological assessment approaches, population and community monitoring) as well as under controlled conditions (e.g. microcosms and mesocosms), while testing under different environmental contexts and exposure scenarios, has allowed us to improve our understanding of the direct and indirect effects of PPPs, their dynamics, and the role of biotic interactions and different environmental factors on these effects. In addition, the development of new *in situ* bioassay approaches (e.g. Afnor standard for monitoring acetylcholinesterase [AChE] enzymatic activity in gammarids; Afnor, 2020) has helped to improve the biomonitoring of ecotoxicological effects, including those of specific PPPs, including within the framework of regulatory approaches.

Furthermore, the development of new approaches and techniques, combined with the evolution of existing ones, has allowed the identification of effects not previously detected due to the absence of suitable methods or their low level of sensitivity. Thus, the development of approaches such as the inclusion of intra- and interspecific genetic variability and its evolution, multi-generation tests, and the development of tools for assessing epigenetic modifications and endocrine disruption have made it possible to advance the assessment of chronic sublethal effects of PPPs (including post-exposure, in order to study the persistence or resilience of these effects). The behavioural and functional consequences of PPP effects (from the individual to the community level) have also received increasing scientific attention.

However, for all of these aspects, knowledge remains very incomplete and several future research objectives have been identified in the literature. These aim in particular to take better account of the complexity of the environmental context, to better characterise the mechanisms of effects (including sublethal effects and their evolutionary consequences),

to assess the degree of specificity of existing indicators for exposing the effects of PPPs and to develop new ones, and finally to take better account of ecosystem functions. This last point aims, on the one hand, to better understand the indirect effects of PPPs that propagate through some of these functions and, on the other hand, to better qualify and quantify the consequences of the contamination of ecosystems by PPPs on their ecological functioning (in order to contribute, in particular, to an improved assessment of these consequences at the ecosystem services level).

## ■ Complexity of the environmental context

The decline in biodiversity is multi-causal due to the multiplicity of pressures (habitat degradation/loss, consequences of climate change, multiple chemical pollutants including PPPs, overexploitation of species, proliferation of diseases and invasive species, etc.), most of which are interdependent. Studies aiming to disentangle the interactions between these different pressures are rare and, as a result, these interactions are not taken into account by current regulatory procedures (EFSA Scientific Committee, 2021).

Moreover, the complexity of the relationships between PPP contamination of the various environmental matrices, exposure of organisms and the resulting effects depend on a range of factors. Some of these include, on the one hand, the type of substance, its time of application and its fate in the environment and, on the other hand, the types of organisms, their life traits, their sensitivity to PPPs (which may vary according to their developmental stage), their capacity to adapt and the biotic interactions they face. These different factors are themselves regulated by the above-mentioned pressures.

Understanding the specific role of PPPs in the decline of biodiversity and improving the regulatory environment for these substances therefore requires consideration of the complexity of the environmental context, including the effects of mixtures and multiple stressors, and the consequences for environmental change (Bruhl and Zaller, 2019; Topping *et al.*, 2020).

Meeting this ambitious objective therefore implies the implementation of improved synergies between the three complementary approaches already used to date: *in situ* studies; studies under controlled laboratory conditions (e.g. microcosms) or semi-experimental conditions (e.g. *in situ* mesocosms or plots equipped with instruments); and modelling approaches.

### In situ studies

*In situ* studies by their very nature make it possible to consider environmental complexity. On the basis of observations carried out at different spatial and/or temporal scales, they favour the use of correlative approaches that allow the establishment of hypotheses on the causal links between the population dynamics or communities observed and the environmental stresses measured (including those caused by PPPs). The robustness of these hypotheses and the degree of generality of the results obtained most often depend on the size of the spatial and temporal scales considered and the amplitude of the environmental

gradients covered (e.g. contamination gradients, climatic gradients). They also depend on the quantity and quality of the metadata used in the analyses.

These different criteria must be considered in order to increase the number and power of field studies aimed at separating the influence of PPPs from that of different environmental factors on a wide variety of organisms, in order to acquire new knowledge and increase the robustness of existing knowledge. To achieve this, it is necessary to select sites with orthogonal gradients of the variables of interest and in which population trends, individual health and demographic performance, and environmental characteristics (e.g. landscape, habitats, pathogenic pressure, agricultural practices, and the application and fate of PPPs) are recorded simultaneously. The establishment of long-term monitoring systems should be based on platforms or systems that already exist at the international and national levels, such as the Zones Ateliers network or the RECOTOX initiative (an ecotoxicology initiative to monitor, understand and mitigate the impacts of pesticides in socio-agroecosystems), but also on monitoring systems set up by various environmental stakeholders involved in epidemiology and ecotoxicological surveillance. This also requires a major commitment to the storage, sharing and processing of data (and associated metadata), which must be organised and supervised at the level of the entire scientific community working on the issue, involving, if possible, the various stakeholders likely to generate and/or use these data.

### Approaches under controlled conditions

Approaches under controlled laboratory or semi-experimental conditions (from microcosms to instrumented experimental sites) complement studies in the natural environment to better identify and characterise the cause-effect relationships between PPP exposure and the biological responses observed at different scales (from sub-individual to community). More relevant from an ecological perspective than laboratory bioassays, they permit the testing of selected PPP exposure scenarios, while addressing varying levels of biological (e.g. populations or communities, one or more trophic levels) and/or environmental complexity (e.g. PPPs alone or in combination, inclusion of other stressors), in order to better understand the mechanisms of biological responses and their potential ecological impacts.

For example, in the context of climate change, these approaches allow for testing the influence of different scenarios (e.g. increase in temperature or in the amplitude of its fluctuations, increase in the frequency and amplitude of extreme events such as droughts) on the effects of PPPs. However, the literature review has highlighted the need for more knowledge on this topic, which is usually addressed by considering only one factor associated with this change (most often a temperature increase).

The effects of mixtures of PPPs also remains understudied to date, even though organisms are usually exposed to different PPPs, whether simultaneously and/or sequentially. Environmental exposure also includes in many cases other types of chemicals in addition to PPPs. These exposures to mixtures of substances can lead to complex synergistic

effects beyond the addition of the effects caused by each substance separately. When it is addressed, the question of the effects of mixtures is mostly dealt with through studies that consider simultaneous exposure to different PPPs, and which do not adequately take into account the temporality of successive exposures. Moreover, as noted by Jonker et al (2005), the experimental designs employed do not always allow a genuine interaction between the PPPs to be demonstrated, beyond a simple additive effect. Finally, there have been very few studies dealing with the influence of adjuvants and co-formulants.

The difficulty in addressing the effects of mixtures and multiple stresses in experimental studies is partly due to the fact that the scientific community still faces conceptual and methodological limitations in dealing with the multiplicity of possible scenarios.

Moreover, this CSA highlights the influence of different types of biotic interactions (e.g. symbiotic relationships, prey-predator relationships, competition, habitat) in the propagation of PPP effects. However, knowledge on this subject remains relatively fragmentary due to the limited consideration of these interactions in experimental studies. Regarding vertical trophic interactions, the biological variables generally measured to assess the impacts of PPPs on food webs are attack and predation rates, consumption rates, survival, abundance and biomass, which do not provide information on the consequences on the health status and physiology of organisms. The latter are more difficult to measure, but provide additional information on the effects that lead to trophic changes. Thus, relatively recent studies tend to favour effects descriptors based on life history traits and behaviour where possible. In the case of aquatic invertebrates, for example, there are studies showing or suggesting PPP-induced increases in vulnerability to predators, which may involve behaviour (movement by drifting with currents) or altered health (muscle mass, reserves). In addition, a few rare studies demonstrate, on the one hand, that PPP effects on mycorrhizae and rhizobacteria can impact plant growth and, on the other hand, that PPP effects on the microbiota can have important consequences for exposed organisms (e.g. alteration of bee gut microbiota by glyphosate; Motta *et al.*, 2018), thus demonstrating the importance of considering the influence of such symbiotic relationships on the response of organisms to PPP exposure.

In general, it would seem important in PPP impact studies to employ sets of indicators that consider different types of organisms (spread over several trophic levels and representing different vertical and horizontal relationships) in order to account for the direct toxicity of substances (according to their mode of action) and indirect effects (according to the characteristics of the system studied).

However, it is important to note that approaches under controlled laboratory or semi-experimental conditions still have limitations. Thus, although these experimental designs incorporate a greater degree of complexity, the validity of the conclusions drawn from these studies remains limited to the conditions and exposure scenarios chosen, which may differ from real conditions. In particular, the spatial and temporal scales considered may be too small for some situations (e.g. changes in landscape or land use) and some biological models (e.g. the largest and/or most mobile organisms, and/or those with life

cycles of several months or even years). The absence, in most cases, of connectivity with the surrounding environment is also a limiting factor in understanding the effects and their dynamics (including those of mitigation or resilience) as they occur in the environment. Thus, the effect thresholds of PPPs found *in situ* are in some cases much lower than in studies carried out in microcosms or mesocosms, which may then lead to an underestimation of ecotoxicological risks. This has been demonstrated in particular in studies using toxic units (from experimental data) for aquatic invertebrates, with threshold differences of up to a factor of 1000 (Schäfer *et al.*, 2012; Liess *et al.*, 2021). This clearly calls into question the relevance of the safety factor values currently applied to determine regulatory thresholds of effects based on toxicity values from laboratory trials.

### Modelling approaches

Modelling approaches based on mathematical tools have been constantly evolving, particularly since the early 2000s. The array of existing ecotoxicological and ecological models have been developed for different species (primary producers, microorganisms, invertebrates and terrestrial and aquatic vertebrates) present across all environments and with different objectives: assessing the sensitivity of species and the vulnerability of certain populations, estimating bioaccumulation and trophic transfer, protecting ecosystem services, etc. The models also allow the estimation of different effect thresholds that can be used in PPP risk assessment (possibly including transformation products, co-formulants, adjuvants and impurities that have not yet been tested) and the extrapolation of the effects of these substances to a large number of species in other types of environments. These models have the advantage of guiding experimental strategies, prioritising assessment scenarios and, if necessary, limiting tests on certain organisms. For example, modelling can contribute to the assessment of the effects of mixtures and, in particular, guide the choice of mixtures to be tested as a priority (e.g. those for which the models predict effects beyond acceptable thresholds in terms of regulation and/or preservation of the environment and its biodiversity), while helping to unravel the mechanisms underlying the observed effects (Belden and Brain, 2018). At the individual and population levels, the use of toxicokinetic and toxicodynamic (TKTD) models applied to the study of the effect of mixtures appears to be a particularly promising approach, as it allows the evolution over time of the exposure of organisms to these mixtures and the resulting effects to be factored in (Bart *et al.*, 2021).

However, even though models of effects on organisms at the individual or population level increasingly integrate exposure to PPPs, this literature review found few models that quantitatively combine these two aspects (exposure and effects). Therefore, the integrated development of fate models (e.g. degradation, sorption, speciation, transfer dynamics) of substances in the environment, bioaccumulation models and effect models at the different levels of biological organisation is desirable if we are to make risk assessment more robust in a realistic environmental context.

In addition, existing models rarely consider chronic sub-lethal or transgenerational effects. They also overlook the effects of the various pressures mentioned above (notably habitat degradation/loss, consequences of climate change, multiple chemical pollutants). For example, there is a need to take greater account of the role of the landscape (e.g. composition, structure, connectivity) in regulating the exposure and effects caused by PPPs. More recent, notable innovations include field studies with the use of models to explore the consequences of different landscape management options on populations. An example of such an approach was recently undertaken in Canada to document the links between landscape-scale agricultural intensification, PPP usage, contamination of insects, and joint declines in insect (prey) and swallow (predator) populations (Garrett *et al.*, 2021; Poisson *et al.*, 2021).

The limited use of population models, whether for forecasting risk or for assessing the impacts of PPPs in natural environments, reflects the lack of formalisation of indicators and common interpretation frameworks, which are more developed in conservation biology, wildlife management or for monitoring epidemic trends in the health field. As for community and food web models, they currently lack the sophistication required to address certain ecological processes, and there remain very few models that combine ecotoxicology and ecology. This can be explained in part by a lack of available data to inform these models. This observation also applies to certain contexts, in particular the marine environment and the French overseas territories, for which the scenarios and data generated by modelling are difficult to use in their current state.

Furthermore, this literature review reveals a lack of sensitivity and uncertainty analyses of these models, the performance and reproducibility of which is rarely tested.

## **I Effects mechanisms, including sublethal effects and their evolutionary consequences**

To understand the mechanisms involved in population and biodiversity declines, it is necessary to combine monitoring of population trends with individual and sub-individual measurements of the parameters determining the individual fitness of organisms that influence population dynamics, in order to better understand the causal links between the trends observed and the pressures to which these populations are exposed. With regard to the specific impact of PPPs, research logically tends to focus on types of effects close to the mode of action of the substance(s) studied, and on species close to the organism targeted by them. This therefore creates a risk of underestimating effects unrelated to the mode of action, including on species that may be very different from the target taxon.

### **Sublethal effects**

An increasing number of studies are highlighting sub-lethal effects at the organism level (e.g. disruption of the nervous, hormonal and immune systems) and even at the holobiont level (disruption of interactions with microbiota). These effects can have repercussions on populations by impacting, for example, growth, reproduction, feeding, predation,

defence (including anti-predation) or orientation. They may result from toxicity mechanisms during chronic exposure at low concentrations that differ markedly from acute toxicity modes of action.

Numerous sub-individual biomarkers can be used to detect the effects of PPPs, for example to determine their genotoxicity (e.g. chromosomal abnormalities, nuclear anomalies, micronucleus testing), their neurotoxicity (e.g. AChE activity) or their effects on immunity (cellular functions such as phagocytosis and non-specific humoral functions such as lysozyme and complement factor activities, or specific functions such as circulating antibody levels). However, translating observed effects to impacts on the health of organisms remains difficult.

The rapidly developing 'omics' tools allow the effects of PPPs to be identified at the molecular and biochemical levels (e.g. metabolic pathways), at different levels of biological organisation, from the cell to the community, via the individual and the population, from (meta-)genomics to metabolomics. For example, transcriptomics is concerned with gene expression and targets mRNA, the intermediate product between genes and proteins. Proteomics measures all of the proteins produced, and metabolomics (particularly lipidomics) is concerned with the metabolites in cells and organisms. The application of these approaches could help to improve mechanistic knowledge of PPP-molecular target interactions by identifying metabolic pathways altered in response to PPP exposure.

These different 'omics' approaches are also of interest when applied in the conceptual framework of the adverse outcome pathway (AOP) in order to understand the mechanisms and cascade effects induced by micropollutants such as PPPs. AOPs are not substance-specific. Rather, they describe an adverse effect pathway from a molecular initiating event (through a cascade of effects). Knowing this effect pathway, an AOP can thus be used to categorise toxicants (if they trigger the initial event, then the adverse effect can be predicted). On the other hand, AOPs are generally organism-specific, so annotated reference genomes are useful for developing new AOPs.

### Evolutionary consequences

The study of sublethal effects would benefit from taking better account of possible evolutionary consequences, which are probably largely underestimated to date, and which raise the question of the physiological or ecological cost associated with adaptation to PPPs. Evolutionary dynamics are impacted by PPPs because of the selective advantage conferred on individuals and species that are more resistant to the toxicity or adapted to the pressure exerted, and because of the genetic or epigenetic modifications induced and transmitted over the generations. At the community level, it therefore represents a response system to the effects of PPPs that itself changes over time, with consequences that are still poorly understood. This is an emerging field of research, with studies conducted until now mainly on short-lived organisms such as microorganisms or invertebrates. Since the end of the 20th century, the rise and development of molecular methods have made it possible to more precisely explore the effects of PPPs on the structure and

diversity of microbial communities using molecular fingerprinting analyses and studies of the diversity of the amplicon sequences of different genes or intergenic regions (ITS) to investigate the diversity of organisms such as prokaryotes (16S rDNA for bacteria, cyanobacteria and archaea) or eukaryotes (18S rDNA for microscopic fungi and microalgae or fungal ITS for the former). The analysis of these markers makes it possible to determine the impact of PPPs on the alpha (e.g. Shannon index) and beta diversity or the relative abundance of these different taxonomic groups within microbial communities. The emergence in recent years of metabarcoding approaches has enabled the data generated by sequencing approaches to be better exploited and community diversity to be better understood. Applied to benthic diatoms (microalgae), this technique has notably shown its potential for estimating the ecological quality of rivers (Vasselon *et al.*, 2017). These molecular approaches also make it possible to explore the effects on intraspecific diversity. Population genetics, which was born a century ago, provides a valuable and still growing body of theory, models and markers specifically dedicated to the study of this level of diversity. This discipline describes and analyses the distribution of genetic (nucleotide) polymorphism within and between populations of the same species, as well as its evolution over time, in order to infer the evolutionary forces at work (e.g. genetic drift, selection, reproductive systems, gene flow) that are responsible for the observed patterns of variation. Current high-throughput sequencing techniques have more recently led to the development of population genomics, which, thanks to its 'omics' dimension, estimates genetic diversity along genomes with increased resolution and makes it possible, for example, to look for selection signatures between or within differentiated populations. These new tools can be particularly useful in the ecotoxicological context, when PPP pressure is suspected of having a selective effect. Moreover, the acquisition of new genomic resources (microsatellite markers, nucleotide polymorphisms, whole genomes) is continuously and exponentially increasing (e.g. public databases, National Center for Biotechnology Information, or NCBI, European Bioinformatics Institute, or EBI). These resources offer the possibility of studying the micro-evolutionary processes (short times, demogenetic approaches) resulting from this type of pressure in species selected for their ecological relevance. The acquisition of new reference genomes (e.g. initiatives such as i5k, or Sequencing Five Thousand Arthropod Genomes; Poelchau *et al.*, 2015) makes it possible to extend the field of comparative genomics and molecular evolution. It is also conducive to the development of AOP approaches (e.g. validation of in vitro molecular models adapted to model species selected on ecological criteria). Finally, molecular and environmental epigenetics has the potential to distinguish processes involving phenotypic or developmental plasticity vs. genetic adaptation in the true sense.

Linking such approaches to laboratory toxicity experiments and experimental evolution should prove particularly informative for understanding the evolution of genetic adaptations in the context of chronic exposure of natural populations to PPP-related stresses (e.g. case of multiple evolutions of pyrethroid resistance in a freshwater crustacean; Weston *et al.*, 2013).

The development of this research into evolutionary and adaptive phenomena would require the consolidation of studies incorporating post-exposure monitoring, an understanding of the influence of successive/repeated applications on resilience capacities, and better consideration of the transgenerational consequences as well as the possible physiological or ecological costs that may be engendered by these phenomena.

## ■ Specificity of indicators

Given the multiplicity of factors influencing the observed biological responses and their interactions, establishing the proportion of effects specifically attributable to exposure to PPPs is a major challenge.

The importance of this issue is fuelling a growing interest in effects-based approaches to regulatory monitoring of ecological quality. At the community level, indicators based on the study of biological and ecological traits, such as the SPEAR (species at risk) method, or those based on the study of the tolerance capacities of communities, such as the PICT method, can be cited. The SPEAR approach was developed in the early 2000s from the study of benthic macroinvertebrate communities (Liess and von der Ohe, 2005), which enabled the impact of PPPs on this type of community to be documented in various rivers, including some in France. The PICT approach has been applied to microbial communities for more than thirty years (Bérard *et al.*, 2021). These two methods, which have strong operational potential, particularly for monitoring the quality of aquatic environments in a context of PPP contamination, have for example been successfully used to specifically highlight the impact of herbicides in rivers on benthic diatom communities (Pesce *et al.*, 2016; Wood *et al.*, 2019).

As mentioned above (see 'Effects mechanisms, including sublethal effects and their evolutionary consequences' section), many biomarkers enable the detection of PPP effects on organisms. However, in the context of *in situ* monitoring, interpretation is hampered, as is the case for many observed effects, by the non-specificity of most of these biomarkers and by the influence of confounding factors that are not always accounted for (other pollution and environmental stresses). The implementation of calibrated *in situ* bioassays can partially offset this last constraint (e.g. Afnor AChE standard; Afnor, 2020).

In addition, it is also necessary to develop new biomarkers and specific indicators at different biological scales. Increased commitment to research and the combination of 'omics' approaches to understanding ecotoxicological responses could help to achieve this objective, for example by targeting genes involved in resistance or biodegradation mechanisms specific to certain PPPs. Modelling approaches based on 'omics' type data are still lacking, although they appear to offer genuine potential for PPP risk assessment (e.g. detection of early effects).

Ultimately, the combination of specific indicators and more traditional ecological indicators would make it possible to better qualify and quantify the role of PPPs in the general decline of certain populations at the European scale (e.g. insects, farm birds, amphibians,

bats), or possibly even to detect other effects on other populations whose diversity and/or sensitivity to currently-used PPPs is still poorly known. Such an orientation brings strong data and associated metadata challenges with regard to the compilation, sharing and processing of information in order to be able to make the best possible use of the scientific and possible operational outputs of this type of multi-indicator approach.

## **I Consideration of ecosystem functions**

Many studies highlight the impact of PPPs on a wide range of ecosystem functions, generally understood from functional descriptors associated with different biological activities (e.g. organic matter degradation, photosynthetic activity, biodegradation, bioturbation). However, some functions are almost never addressed in terms of PPP impacts (see 'Impacts on ecosystem functions' section). This is the case, for example, for functions relating to the retention and regulation of water flows in soils and sediments, albedo, and the formation and maintenance of soil and sediment structure. Yet the reported effects of PPPs on biodiversity and on a wide range of biological activities and ecological processes involving various organisms in terrestrial and aquatic environments, in particular plants, micro-organisms and invertebrates, are likely to impact on these functions.

For example, it is likely that the effects of herbicides, by acting on the photosynthetic microbial biomass on the soil surface, the physiology and diversity of primary producers, or the root system, indirectly impact the retention and regulation of water flows as well as albedo and reflection through changes in aerial plant cover, with potential consequences for soil fertility. They can also affect the anti-erosion functions of primary producers, with potential consequences for the transfer of PPPs themselves, as well as for coastal and riverbank erosion, flooding and mudflows, all of which are increased by climate change.

Bioturbation (a process of soil movement by animals that directly or indirectly affects soil and sediments, and includes both particle reworking and the breakdown of burrows) is another ecological process for which PPP effects have been documented. For example, several studies have shown impacts from copper on the structure of contaminated soils as a result of a decrease in earthworm burrowing activity. Such effects may have consequences on water retention and flow regulation functions as well as on the maintenance of soil and sediment structure, which may impact on agricultural production.

These observations show the need to move beyond the study of impacts on biological activities and ecological processes by developing approaches based on the concept of ecosystem function. From this perspective, it could be useful to develop impact indicators based on the functional traits of organisms, paying particular attention to species and communities considered to be ecosystem engineers due to the strong influence of their activity on the physical structure of habitats. It would also be relevant to better harness functional indices such as the rate of litter degradation by microorganisms and invertebrates or pollination measurements. Finally, it is also important to better understand the role and limits of functional redundancy in the mitigation of PPP impacts. Broadly

speaking, concepts and tools from ecology (e.g. ecological indices, network theory, and ‘omics’) could be more widely used to assess the functional impacts of PPPs.

In addition, a particularly innovative approach would involve coupling ecotoxicological models with models that provide information on the links between the species present and the functions they perform within ecosystems, as well as the services associated with them.



## 4. Consequences for ecosystem services

The complexity of the interactions in which pressures from PPPs and their evaluation has been shown in the previous sections. For PPP risk management, decisions are also based on difficult trade-offs between desired vs. undesired effects, taking into account the question of the acceptability of effects, depending on their magnitude. The difficulty of informing decisions on the basis of anticipation of ecological processes has favoured the emergence of a risk assessment approach incorporating the concept of ecosystem services. This instrument, which is understood to integrate a range of dimensions, is intended to improve the clarity of the consequences of PPP use on the benefits derived by humans from biodiversity. At the European level, EFSA in 2010 published recommendations for the consideration of ecosystem services within the definition of specific protection objectives in the context of European legislation on PPPs (EFSA Panel on Plant Protection Products and their Residues, 2010). This work subsequently formed the basis for the development of guidelines to better protect biodiversity and ecosystem services from the adverse effects of PPPs or other contaminants (EFSA Panel on Plant Protection Products and their Residues, 2013; Benford *et al.*, 2016). However, scientific articles published on this subject over the last decade show that the operational methods for harnessing the concept of ecosystem services for chemical risk assessment have not yet been established.

In line with the work carried out by EFSA, many scientific articles have discussed the inclusion of ecosystem services in the assessment of the effects and risks of chemical contaminants. In particular, some authors have highlighted the possibility of assessing the relationship between the benefits provided by the use of PPPs on the one hand, and the environmental costs resulting from the contamination caused by this use on the other hand. There is therefore a school of thought that considers that the ecosystem services approach to the development and implementation of environmental risk assessment procedures is the best strategy for strengthening the ecological dimension of environmental regulation, by emphasising the benefits of nature protection. However, there is also a consensus within this school of thought that the application of this approach in the context of PPP regulation still faces many scientific and methodological challenges. Furthermore, a section of the scientific community is questioning the validity of approaches centred exclusively on the concept of ecosystem services or, more broadly, on the concept of the contribution of nature to the benefit of humans, which is sometimes associated with a simplistic and utilitarian vision of the environment. In view of this questioning of the sometimes very real conflict between nature protection *per se* vs. human benefit, the IPBES conceptual framework recognises the intrinsic value of nature (Diaz *et al.*, 2015).

In order to analyse the impacts of PPPs on biodiversity by identifying the resulting consequences on ecosystem services, the links between, on the one hand, the effects of PPPs on ecological processes and functions identified in the ecotoxicology field and, on the other hand, effects on ecosystem services are first clarified at the conceptual level in order to agree on a common frame of reference for the CSA. The results of the most significant existing assessments of the consequences of PPP use on ecosystem services are then summarised. This analysis leaves the field of investigation wide open, and the main avenues that emerge are discussed.

### Conceptual links between functions and services

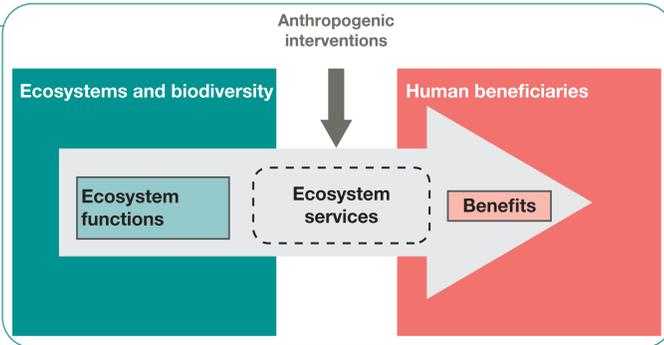
The concept of ecosystem services emerged in the 1970s. It was first used by economists to conceptualise the link between the functions of nature and the benefits that society derives from them. Subsequently, major works such as those by Daily *et al.* (1997) and Costanza *et al.* (1997) gave a multidisciplinary dimension to this concept, and proposed the following definitions: "the conditions and processes through which natural ecosystems and the species that make them up sustain and fulfil human life" (Daily *et al.*, 1997); "the benefits that human populations derive, directly or indirectly, from ecosystem functions" (Costanza *et al.*, 1997).

The concept of ecosystem services has henceforth been used by international bodies and partnerships as an instrument to promote a better awareness of the consequences of biodiversity loss and to facilitate the guiding and coordination of initiatives by integrating the multiple dimensions of biodiversity into this common reference framework. Based on these initial definitions, the concept has been the focus of a series of initiatives within the framework of the UN or international research programmes, including the Millennium Ecosystem Assessment (MEA) in 2000-2005, and The Economics of Ecosystems and Biodiversity (TEEB) in 2007-2011. These efforts to synthesise knowledge led to the creation in 2012 of the Intergovernmental Platform on Biodiversity and Ecosystem Services (IPBES). The main objectives of these initiatives were, respectively, to consolidate the conceptual framework of the ecosystem service concept (MEA), to estimate the value of these services in economic terms (TEEB) and to create, on this basis and in the same manner as for climate with the IPCC (Intergovernmental Panel on Climate Change) an interface between scientific expertise and governments on issues relating to biodiversity and ecosystem services, in order to implement the necessary strategies for their conservation and sustainable management (IPBES).

As part of this trend, the EU has implemented the Mapping and Assessment of Ecosystems and their Services programme, and in 2012 France initiated the French Assessment of Ecosystems and Ecosystem Services (Efese). Efese brings together a range of assessment studies on ecosystems and their services at different scales, in order to support the *Stratégie nationale pour la biodiversité* (National Biodiversity Strategy) and contribute to the achievement of the Sustainable Development Goals.

Efese first defined ecosystem goods and services as "socio-economic benefits derived by humans from their sustainable use of the ecological functions of ecosystems" (Efese, 2016). This definition was then further developed in the context of the valuation of agricultural ecosystem services (Tibi and Therond, 2017) to further clarify the distinction between ecological functions, ecosystem services and benefits derived from an ecosystem service by one or different stakeholders, in monetary or non-monetary form (Figure 16). This distinction is important because different ecosystem functions can contribute to the provision of an ecosystem service, and one function can contribute to different services.

**Figure 16. Constituent elements of an ecosystem service according to the Efese conceptual framework (after Efese, 2016)**



This conceptual framework was chosen for this CSA, based on the recently updated Common International Classification for Ecosystem Services (CICES, version 5.1; Haines-Young and Potschin, 2018). It structures ecosystem services into three categories: provisioning services, regulating services and cultural services, with regulating services underpinning the proper functioning of the other two categories.

Based on this framework and the one established for functions (see 'Impacts on ecosystem functions' section), 17 experts within this CSA representing a variety of disciplines (environmental chemistry, agronomy, microbial ecotoxicology, aquatic ecotoxicology, terrestrial ecotoxicology, ecology and evolution, fate and effects modelling) were approached to identify potential direct and indirect links between each group of services and each category of functions.

In summary, it is evident that all groups of ecosystem services potentially rely on all categories of functions. In particular, a majority of experts in the panel considered that 95% of the combinations of provisioning services and regulating and maintaining services are characterised by direct and indirect links to the different categories of ecosystem functions that have been proposed (this is less the case for cultural services, for which most experts and scientific found it difficult to express an opinion). The analysis also revealed that the perception of the nature of these links may differ according to the experts' disciplines.

The experts also emphasised the variable nature of the relationship between ecosystem functions and services, which can be positive or negative, or of low or high magnitude, depending on the time scale and the context in question. Moreover, few services appear to be linked to only one category of function. This suggests that prioritising the services to be preserved would not ultimately restrict the range of functions or processes to be considered.

## Principal ecosystem services impacted

Within this subject area, marked disparities are apparent in terms of available knowledge. Regarding environments, the terrestrial environment has been the subject of most of the studies, while little information is available on continental and marine aquatic environments. With regard to ecosystem services, four are the subject of particular attention in relation to the impact of PPPs: food production, biological control, pollination and maintenance of water quality. Water quality is most often addressed in terms of impacts on human health and clean-up costs, and has therefore not been considered here. The same applies to PPP contamination of food, with consequences for human health and the market value of food, which are outside the scope of this CSA. By contrast, the provision and maintenance of soil quality is receiving increasing attention, despite the fact that there has been little research on the impacts of PPPs on soils. Finally, cultural services are rarely studied.

## Provisioning services

The provisioning service for crops is the most extensively studied service. Its relationship with ecosystem functions is complex. Crop yields result from primary production through photosynthesis and from the functionalities provided by regulating services (e.g. soil quality and nutrient supply, water regulation, pollination, pest regulation), but they are also highly dependent on human inputs. In particular, PPPs are used in the production process to eliminate a disservice (understood as a disadvantage to humans from ecosystems and biodiversity), namely the action of crop pests. However, this same disservice is sometimes seen as being favoured by the manner in which crops are grown (lack of diversity, and use of PPPs in particular which in response generates an increase in pest populations).

While highlighting the expected positive short-term impact of PPP use on provisioning services, particularly food services, due to their action in protecting crops from the disservice of pests, scientific articles suggest a negative longer-term impact of PPPs on these ecosystem services. This notion of temporality has yet to be substantiated. The research by Deacon et al (2015; 2016) estimating the impacts of insecticide use on the maintenance of the provisioning service in lemon cultivation in Spain and tomato cultivation in Italy, respectively, underlines the value of a careful use of PPPs in combination with other practices, such as the establishment of protected vegetation zones, at

time frames of ten to fifty years. Fisheries is also a provisioning service that can be negatively affected by PPPs, as demonstrated by De Valck and Rolfe (2018) in their work on the impacts of three major types of pollution (nutrients, sediment deposition and PPPs) in three major Australian coastal ecosystems (mangroves, seagrasses and coral reefs).

The use of economic valuation approaches that include the concept of consumer surplus, i.e. the fact that consumers are prepared to pay more for certain types of products or services, shows that there is a social demand for agricultural products that are produced using more environmentally friendly practices.

Although there are exceptions, the literature analysed in this CSA highlights a tension between crop production on the one hand and other services on the other. Thus, the overall message from the literature reviewed is that agricultural practices should aim to minimise the pressure of PPPs on biodiversity and ecosystem services.

### ■ Biological control

Biological control, which is defined as the pest control service provided by natural enemies, is also one of the most studied ecosystem services because of its importance for agricultural production. A body of work agrees that the exclusion of natural predators, particularly through the use of PPPs, will lead to an increase in the presence of pests in crops that can be substantial. However, as PPPs are used in combination with other practices that also impact natural predator populations (e.g. plot expansion, habitat destruction), their contribution to the degradation of the biological control service is difficult to establish in isolation.

As with pollination, this work highlights the importance of maintaining natural or semi-natural habitats to preserve the biological control service. Reducing the use of PPPs and maintaining natural predators through these habitats would reduce production costs and increase the social benefits associated with the crops under review.

### ■ Pollination

The pollination service provided by bees, and more widely by pollinating organisms, is one of the most intensively studied ecosystem services. Entomophilic pollination is indeed essential to certain agricultural crops, and its value has been the subject of monetary valuation. Today's leading assessments put the value of pollination services at between 153 and 422 billion US dollars for the year 2005 at the global level (Gallai *et al.*, 2009), an order of magnitude confirmed by the IPBES in 2019 (IPBES *et al.*, 2019) where the value was between 235 and 577 billion US dollars for 2015. At the French national level, Efese<sup>21</sup> provides a range of 2.3 to 5.3 billion euros per year for the period 2006-2010. This regulation and maintenance service strongly interacts with the food production service,

21. <https://www.ecologie.gouv.fr/sites/default/files/Th%C3%A9matique%20-%20Efese%20-%20Le%20service%20de%20pollinisation%20-%20Analyse.pdf> (accessed 9/01/2023)

since it can improve not only the yield but also the quality of agricultural products (e.g. appearance, nutritional quality, shelf life).

Several studies conducted in different regions of the world and on different crops show that the use of insecticides (in particular neonicotinoids) affects pollination due to the direct toxicity of these substances on pollinators (see 'PPPs are a major contributor to the decline of certain taxonomic groups' section). These results indicate that the use of PPPs should be reduced in the field, as the benefit to pollinators would exceed the benefit of PPPs. The use of herbicides has also been documented as impacting pollination by reducing the resources for insect pollinators, and therefore the number of visits to crops. Thus, the presence of natural habitats or the establishment of hedgerows are beneficial to pollination by improving food resources and habitat, and thus the density as well as the diversity of pollinators.

Pollination is an ecosystem service of great concern to consumers, and one for which consumers can change their purchasing behaviour by agreeing to pay more for products certified as better for bees (Wei *et al.*, 2020).

## **I Regulation and maintenance of soil quality**

With regard to the regulatory and maintenance services associated with soils such as carbon sequestration, water storage, fertility and nutrient supply, as well as pollutant uptake and degradation, the results agree that excessive use of PPPs will lead to degradation of soils along with most of the services they provide. Studies comparing different plant protection practices converge to show that PPP-free soils are of better quality, and are characterised by greater microbial biomass and diversity and greater abundance of soil fauna, especially earthworms. However, it is necessary to increase knowledge, firstly, of the effects of PPPs on the ecological quality of soils by promoting work on the ecosystem functions associated with this compartment and, secondly, on the consequences that these effects have on the ecosystem services provided by soils.

## **I Cultural services**

The impacts of PPPs on cultural services are poorly studied, and mainly concern landscape amenities, tourism and recreational fishing. Services are assessed using stated or revealed preference approaches to capture the values of aesthetic and heritage services and recreational services. Other estimates include lost profits from tourism and recreation activities. However, the economic literature generally shows that the share of cultural services in the total economic value is often significant, including for agroecosystems. Studies assessing the impact of PPP use on cultural services would therefore be of interest.

With regard to non-agricultural areas, the results appear to vary according to the type of area. The benefits to human well-being derived from the richness of biodiversity in gardens and walking areas are generally negatively impacted by the use of PPPs. On the other hand, when cultural expectations are still strongly associated with the strict

control of vegetation, such as in cemeteries, sports grounds or golf courses, cultural services are positively associated with PPPs. However, this benefit associated with PPPs is tending to diminish as cultural expectations shift toward the preservation of ecosystems and biodiversity.

## Innovations and future prospects regarding ecosystem services

### ■ Disconnected research fields

Although the concepts are linked, the literature dealing with the impacts of PPPs on ecological processes and ecosystem functions, mainly within the ecotoxicology field, appears to be disconnected from the literature on ecosystem services. Within the ecosystem services literature, the impacts of PPPs are rarely studied. When they are, the use of PPPs is most often examined in general terms. Few publications mention a substance or family of substances, with the exception of neonicotinoids.

Some authors highlight this lack of connection between the objects of study and criteria for assessing the effects of PPPs within the ecotoxicology field, and the approaches developed for assessing the impact on ecosystem services. Exceptions exist, however, such as a study based on an economic and ecosystem modelling approach to explain the consequences of changes in the relationships within a species network that includes invertebrates (in particular the zoo-phytoplankton relationship) in a lake exposed to organophosphate insecticide contamination, in terms of water purification and the associated ecosystem services, particularly tourism (Galic *et al.*, 2019).

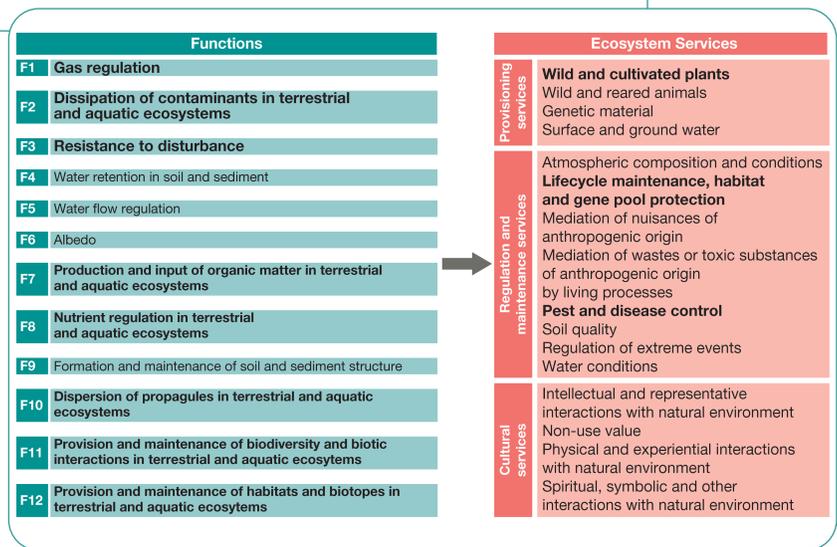
The impact of PPPs on some ecosystem services appears to be relatively undocumented, even though these are related to functions for which the impact of PPPs has been highlighted in the literature. Figure 17 shows the relatively small range of services for which the impacts of PPPs have been studied, compared to the diversity of functions for which effects are reported.

For example, the ecosystem service of developing and maintaining soil quality is linked, among other things, to the activity of primary producers, micro-organisms and terrestrial invertebrates. The extent of the effects of PPPs on these communities, as shown in the literature reviewed elsewhere in this CSA, indicates this to be an area worthy of further study, particularly in relation to impacts on the agricultural production service.

Except for the example of fishing in Australian coastal areas mentioned previously, the marine environment has also not been considered, despite the importance of the effects highlighted, particularly on coastal seagrass beds, mangroves and corals, whose ecosystem functions are highlighted but with no information on the ecosystem services that they support. The high level of contamination of certain fishery resources (bivalves,

certain fish due to bioaccumulation processes) also points to an impact that should be assessed in terms of the corresponding provisioning service.

**Figure 17. Links between ecosystem functions and services** (adapted from Pesce *et al.*, 2023a)



List of functions (left, in the green table) and ecosystem services (right, in the salmon table). In bold: the most frequently documented PPP-related functions and services in the corpus.

## Assessment methods

As regards assessment methods, the vast majority of articles use the market valuation approach to ecosystem services, focusing first on the relationship between PPP use and the level of the food supply service, and then possibly extending to other services useful for agricultural production. Assessment using non-market methods is rare. Although non-market methods were originally developed to address the valuation of cultural services, they are ultimately used as much to complement the valuation of the agricultural production service as they are for cultural or regulatory services.

Cost-benefit analyses, which consist of comparing different scenarios or management options on the basis of economic profitability criteria, tend to neglect many sources of benefits, particularly non-market, and therefore do not yet make it possible to integrate all ecosystem services. Cost-effectiveness analyses, which identify the best option for achieving an *a priori* defined objective that may differ from profitability, are very poorly represented in the literature. However, in principle, they are the preferred method for comparing scenarios, as was done, for example, for water quality.

The literature highlights the complexity of interactions between services, which may be antagonistic or synergistic, and present different implementation and valuation dynamics depending on the stakeholders, scales and timeframes considered. The question of trade-offs between different services is often raised. The concept of a package of services has been proposed to incorporate these interactions into the study objective as much as possible.

### ■ Application to risk assessment

With regard to the use of the ecosystem service concept in risk assessment, the need to identify the main taxa or communities involved in the provision of the various ecosystem services through the ecosystem functions impacted by PPPs, and to define quantifiable indicators to translate the effects of PPPs into an assessment of the consequences of these effects on ecosystem services, have been highlighted. It is also necessary to establish a reference framework to define the levels of effects regarded as acceptable or unacceptable, as well as the corresponding levels of ecosystem services. Strategies to be developed to allow for site-specificity and landscape-level assessment are also mentioned.

### ■ Interdisciplinary connections

A disconnect has been noted between the scientific communities involved in assessing the impacts of PPPs on biodiversity and ecosystem functions, on the one hand, and on ecosystem services, on the other. They have their own objectives but also and more importantly their own approaches and methods. The identification of this barrier requires a better mutual understanding of the concepts of ecotoxicology and ecosystem services.

Finally, as the value of ecosystem services is directly linked to their perception, a better understanding of society's expectations regarding the services impacted by PPPs and of their changes should be the subject of sociological and anthropological research. Similarly, farmers' perceptions of the ecosystem services they use and on which they have an influence remains poorly documented in the French context.



## 5. Cross-cutting areas of concern or improvement

Examining the various steps in the causal links between PPP use and their effects on biodiversity and ecosystem functions and services allows us to identify the cross-cutting areas of concern or improvement that emerge as key elements in the body of literature analysed.

Among the characteristics of the substances, some appear to be particularly decisive in their effects, which raises questions about the choice of products used in crop protection. The physico-chemical transfer dynamics of these substances lead, for some, to accumulation processes that are difficult to predict, and which are observed at the scale of the land-sea continuum. Similarly, the dynamics of the propagation of biological effects and their combination with other stress factors lead to an increase in the vulnerability of ecosystems. Conversely, improvements in contamination levels for certain substances or environments have been demonstrated over the last two decades. However, although progress has been made in the collection of data, scientific difficulties persist in addressing the contribution of PPPs to the vulnerability of ecosystems in a comprehensive manner.

### Issues related to the choice of substances

PPP's are developed to control pests and pathogens in crops. Their effects on living organisms are therefore the fundamental reason for their use. Progressively, the growing attention paid to the preservation of biodiversity has led to the search for a compromise between desired effects on target organisms vs. undesired effects on non-target organisms, using a risk/benefit approach (see 'Targeted, non-targeted or unintended effects' section). In this context, certain characteristics such as CMR toxicity, persistence and susceptibility to bioaccumulation, or lack of specificity, have led to the gradual withdrawal of some of the most concerning substances. However, the replacement of a substance by another whose efficacy is based on other characteristics has proved, in various cases, to generate other effects on non-target organisms.

### Persistence and bioaccumulation

As indicated in Chapter 3, the families of substances for which the effects on biodiversity are the most documented are also those for which hindsight is possible, as these are the oldest and most persistent. They include, for example, organochlorines (e.g. DDT, lindane) and organophosphates (e.g. dichlorvos, methyl parathion) and their transformation

products, as well as triazines (e.g. atrazine, simazine, terbutryn) and phenylureas (e.g. diuron, isoproturon). Most of these substances have been banned from use, but their presence in all environmental compartments and in biota, where some of them accumulate even though their concentrations are gradually decreasing, is still widely confirmed by monitoring systems.

Some substances still in use, such as copper, do not degrade. Copper is a naturally occurring element and is used as a fungicide. However, repeated use over time leads to accumulation in soils and aquatic environments at concentrations that in some cases reach levels that affect organisms. These effects have been verified at sublethal levels for copper, with consequences in particular on primary production (microalgae and cyanobacteria), the decomposition of organic matter and microbial or plant-microorganism interactions. Less persistence is therefore generally sought, but this does not systematically translate into less ecotoxicity.

### **I Efficacy and toxicity**

The quantities of active substances applied should be considered in relation to the weight of these substances.

Progress has been made regarding the efficacy of substances at low or even very low doses. However, they are also potentially toxic at low doses for non-target organisms, and their presence in ecosystems can produce effects that have been documented in the literature. Moreover, their monitoring is made more difficult by the low concentration levels at which they are present, which limits the ability to detect them. Laboratory studies (Thomson and Hoffmann, 2006) have allowed the establishment of PPP toxicity indices, on the basis of which cumulative toxicity values from a set of applied PPPs can be calculated. These values could be taken into account by users in order to determine their choice of products in order to limit the impact on non-target organisms. In particular, it has been shown for a set of 12 active substances used in orchards that high cumulative values strongly reduce the populations of crop protection organisms, especially earwigs (McKerchar *et al.*, 2020).

### **I Intensity and frequency of usage**

Some substances were initially perceived as having relatively little impact compared to others because of their lower persistence and/or ecotoxicity, such as glyphosate and S-metolachlor among herbicides, or neonicotinoids among insecticides. This has resulted in widespread use, which raises other problems linked to the size of the areas involved and/or the repeated nature of applications on the same areas, which leads to the phenomenon of pseudo-persistence of these substances in the environment, i.e. a persistence linked to the fact that the substance does not have time to be degraded between applications. This phenomenon of pseudo-persistence, which concerns the vast majority of environments, limits the possibility of refuge and recovery for non-target organisms.

It also leads to the deterioration of trophic resources and habitats exposed to the substances. Such a phenomenon is documented on a broad scale for substances sold in the largest quantities, but it can also characterise a particular local situation depending on the practices in place.

### ■ Selectivity of the mode of action

A selective mode of action, in theory, allows the actions of the PPP to be focused on its target, and thus limit undesirable effects on non-target organisms. However, this approach has limitations due to exposure pathways or unexpected effects.

While neonicotinoids were initially thought to be highly selective for insects due to their particular affinity for nicotinic receptors, effects on many other taxa have subsequently been identified. Birds are now showing increasing evidence that questions this selectivity, and appear to be more sensitive to neonicotinoid toxicity than other vertebrates (Mineau and Palmer, 2013). The acute toxicity of these insecticides has been underestimated by a factor of 10 for certain wild bird species, compared to the toxicity found in model species (Mallard and Bobwhite quail). Sublethal effects have also been shown to be linked to neonicotinoid-induced nervous system disturbances, with particular consequences for flight ability and migration. These direct effects on birds have, in particular, been documented following the ingestion of treated seeds left on the soil surface.

Furthermore, selectivity of the mode of action does not consider the indirect effects that result from the elimination of the target population. For example, the weed flora controlled by herbicides provides a diversity of trophic resources and habitats for many invertebrates and soil microorganisms. It should be noted that this question of indirect effects resulting from the suppression of the target population does not depend strictly on the selectivity of the substance's mode of action nor on its toxicity, since the same indirect effects can be induced by other pest control methods. It does however depend on the degree of pressure exerted on the target population.

### ■ Alternatives and shifting of effects

The observation of undesirable effects of a substance should, in principle, lead to its abandonment, and possibly its replacement by another substance whose effects assessed *a priori* within the regulatory framework are weaker. However, experience has shown that this substitution may be accompanied by a shift of effects to other environments or other organisms. For example, the use of prosulfocarb to replace phenylurea herbicides, banned mainly because of their persistence in water, has shifted the problem to airborne transfer. Similarly, the pyrethroids intended to replace neonicotinoids require repeated applications and are found in birds, mammals, reptiles and amphibians. They cause various effects, such as individual effects in reptiles and amphibians (life traits, behaviour) and sub-individual effects in mammals (endocrine disrupting effects). Therefore, Grimonprez and Bouchema (2021) emphasise that “the concept of alternative should be

thought of as the set of methods and practices to be deployed at the plot or farm level allowing for a comparable control of the phytosanitary risk”, beyond the simple substitution of one substance by another.

## ■ Biocontrol

Biocontrol substances and organisms (or agents) are considered by public authorities as solutions to be promoted for plant protection. These solutions are partly based on the principles of biological control, which aims to regulate pest populations by introducing biocontrol agents that are antagonistic to them. In addition to the incentives introduced in the mid-2010s to encourage their use (e.g. tax provisions, simplified assessments, exclusion from national PPP use reduction targets), they have been the subject of a national biocontrol deployment strategy (*Stratégie nationale de déploiement du biocontrôle*) since 2020. The French definition of biocontrol PPPs is included in the Rural and Maritime Fishing Code (CRPM, article L.253-6), and includes:

- macroorganisms (insects, nematodes or mites that may be indigenous or non-indigenous), used mainly as insecticides;
- microorganisms (viruses, bacteria or fungi), used mainly as fungicides and insecticides;
- semiochemicals, such as pheromones and kairomones (primarily synthetic), used mainly against insects;
- products containing natural substances of plant, animal, microbial or mineral origin, which can have a wide range of uses.

The literature review revealed that little is generally known about the impact of biocontrol solutions on biodiversity and ecosystem services, and few studies compare the impacts of these alternative solutions to those of synthetic PPPs.

The use of living organisms in biocontrol, whether microorganisms or macroorganisms, brings with it a unique dimension compared to the use of synthetic PPPs. They can in fact multiply, move and colonise other environments. The persistence of these organisms in the environment can sometimes be questioned due to the variability of the biotopes under investigation, with very different environmental parameters from one case to another. For example, in the case of *Bacillus thuringiensis* (Bt), persistence can range from a few days to several years. Due to the persistence of microorganisms, their impact on the soil microbiota has also been the subject of specific publications, mostly concerning bioinsecticides (e.g. Bt) and biofungicides (e.g. *Clonostachys rosea*, *Bacillus subtilis*, *Trichoderma atroviride*). More generally, the few results concerning microorganisms reveal their impacts to be mixed: they can be deleterious (mainly altering biodiversity and the ecological balance of the soil) or beneficial (biocontrol of phytopathogenic microorganisms). Finally, no publications documenting the adaptation of target organisms to the chemicals produced by microorganisms, including antibiotics, were found. In the case of macroorganisms, persistence is fairly well studied, at least in the short term, as it is an essential component of their efficacy. Long-term persistence (more than one year) remains poorly understood. The use of predatory macroorganisms is one of

the biocontrol solutions for which the impact on biodiversity has been the most studied, notably in the case of the harlequin ladybird *Harmonia axyridis*: this species has led to a decrease in the biodiversity of native ladybird species and to the establishment of introduced populations in the biotope.

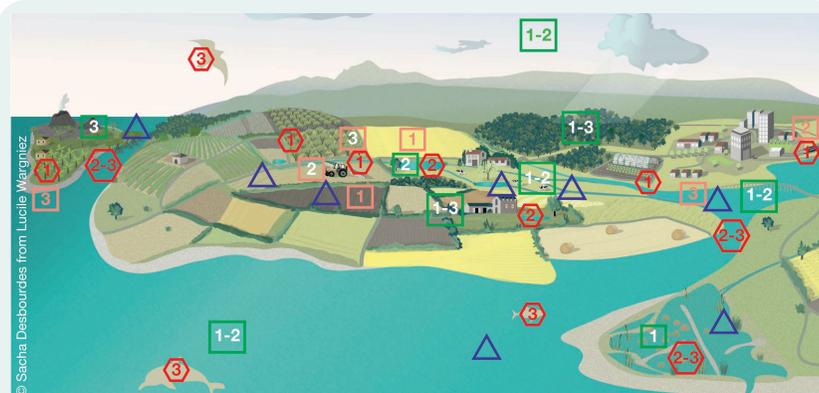
The impact of natural substances is also poorly studied. In general, they tend to have low ecotoxicity. However, some substances (the insecticides abamectin and spinosad) have an ecotoxicity greater than or equal to that of their synthetic counterparts with the same mode of action and molecular targets.

To our knowledge, no study has looked at the effects of semiochemicals on biodiversity and ecosystem services.

It therefore appears that the development of biocontrol alternatives requires more study of their undesirable effects, which at this stage is particularly lacking, to ensure that their use is compatible with the preservation of biodiversity. Recent studies (Jack *et al.*, 2021) have questioned the risk of microbial invasions in agriculture and have warned of the resurgence of the use of microorganisms as biocontrol products, but also as biostimulants. They also question the production and mass release of beneficial microorganisms that could become invasive, with unexpected consequences on plant health, soil communities and ecosystem services. Indeed, beneficial microorganisms can become parasitic (e.g. mycorrhiza), can cause changes in the microbiota and favour invasive plants, or can lead to suppressive soils (e.g. *Streptomyces* bacteria and antibiotic production).

## Accumulation phenomena

In the environment, organisms are exposed to complex mixtures of PPPs, including their possible transformation products as well as adjuvants and co-formulants. Exposure may be simultaneous or successive, with possible accumulation when the substance has not degraded (persistent) or is reapplied too frequently (pseudo-persistent, see 'Intensity and frequency of usage' section). In the soils of treated plots, this temporality depends mainly on agricultural practices. On the wider scale of agrosystems, and outside of them, it will mainly depend on the transfer dynamics of the various PPPs, depending on the environments considered and their distance from the source of contamination. In addition to this chemical pressure, there are generally other sources of stress, which may be linked to the destruction of habitats and/or climate change. Thus, the interconnection of environments does not result in a regular gradient of exposure that decreases from the place of application to the ocean. The situations most often described in the literature as being subject to accumulation, whether of substances or effects, are shown in their geographical context in Figure 18.

**Figure 18. PPP accumulation phenomena and situations of vulnerability**


Element represented	Symbol	Legend
Accumulation of substances		1 Repeated applications and persistence of substances
		2 Retention zones
		3 Bioaccumulation
Vulnerable situations		Evolutionary effects
		Synergistic effects of mixtures of substances
		Multi-stress effects (simplified landscapes, climate change)
Aggravating factors		1 Bare, degraded soils (erosion, loss of organic matter), simplified landscapes
		2 Sealed or compacted soils
		3 Previously weakened ecosystems
Mitigating factors		1 Degradation of substances
		2 Dilution of substances
		3 Refuge areas

## Accumulation of contamination by PPPs

In a general sense, contamination levels decrease along the land-sea continuum, in line with the increasing distances in both time and space from the time and place of PPP application. However, this gradient is not regular and systematic. The fate of substances in the environment involves degradation processes, but also involves retention, accumulation and discharge. The co-occurrence of these phenomena may in some cases favour accumulation in certain compartments (more or less distant from the place of application) of combined substances and/or transformation products. The accumulation gradient can thus be described as the result of two opposing trends that combine according to the characteristics of the land-sea continuum:

- processes that lead to a decrease in concentrations: dissipation and degradation, dilution in water, abiotic (e.g. ultraviolet, temperature, pH) and biotic (e.g. microorganisms) degradation, as well as detoxification processes within organisms themselves;
- processes that lead to increased concentrations: transport by air currents that can lead to localised redeposition, retention in sediments and/or organic matter, accumulation in biota or groundwater<sup>22</sup>.

These processes are ambiguous in their character. Indeed, the processes mentioned above as reducing concentrations may also increase the extent of contaminated areas (e.g. air, estuaries, oceans) and therefore of exposed populations, as well as the diversity of substances present (transformation products in addition to parent substances). Similarly, retention processes can temporarily reduce the exposure of organisms by limiting the bioavailability of substances, but when environmental conditions induce the release of compounds (e.g. soil erosion, floods remobilising sediments) or when prey containing PPPs is consumed, exposure will be even greater.

Depending on the persistence of the compounds, transport may be over varying distances, from local, to regional or even continental.

### Soils and sediments

Soils, particularly those used for agriculture, are directly affected by the application of PPPs and represent an area of accumulation of PPPs, although the concentrations and type of PPPs vary greatly depending on the type of crop grown.

In aquatic environments, sediments may also be areas of accumulation of some PPPs, especially the more persistent and hydrophobic ones such as organochlorines, which are POPs. They are usually transferred in particulate form, bound to suspended matter.

### Transition zones

Mediterranean coastal lagoons and semi-enclosed bays such as *bassin d'Arcachon* (Arcachon bay) and *abers bretons* (inlets on the Brittany coast) which lie at the interface between ecosystems, are prone to retaining inputs from their catchment areas and potentially accumulating them. The pressure linked to PPPs is regarded as significant in these environments, where up to forty or more active substances can be found in individual water samples. These mainly consist of herbicides and fungicides which are linked to agricultural activities in the catchment areas. The concentrations are generally higher and the number of substances detected is greater than in coastal water bodies. Estuaries are dynamic transition zones, sometimes characterised by the presence of a muddy layer that can interfere with the fate of substances brought by rivers to the coast. This zone can effectively act as a trap for certain substances with a high capacity for adsorption on

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22. Groundwater is excluded from the scope of this CSA because of its limited interaction with the organisms studied, but it is an important issue with regard to the accumulation of PPPs.

suspended matter (hydrophobic PPPs), or sometimes, for the most hydrophilic substances, act as a simple transition zone, whereby a conservative transfer occurs solely by dilution.

## Biota

The physico-chemical characteristics of certain substances allow them to persist in the tissues of organisms, where they are neither degraded nor eliminated. As a result, concentrations observed in the tissues increase with each exposure episode. In particular, food webs can be a pathway for biomagnification through the consumption of contaminated prey (see 'Contamination of biota and exposure of organisms' section). In this way, in areas far from the application site where the substances have been diluted or dispersed in the environment, they may still concentrate in the tissues of top predators (in particular marine mammals, carnivorous fish and birds of prey, whose habitat is not highly contaminated but which themselves have elevated concentrations of PPPs). This dynamic also affects transfers between ecosystems, for example from water to land through the consumption of contaminated aquatic insects or larvae by terrestrial organisms, from agricultural areas to wilder areas via birds or large predatory mammals, and from coastal waters to open waters via predatory fish.

## I Contexts and situations leading to vulnerability

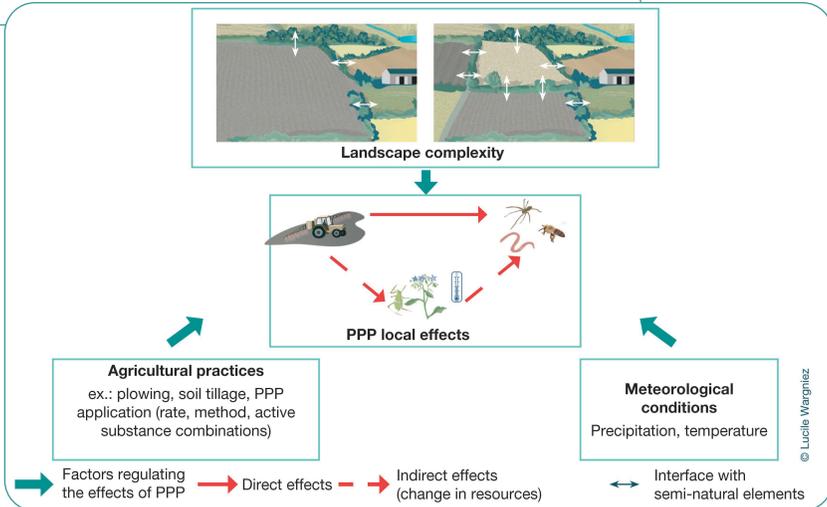
The vulnerability of an ecosystem to PPP use results from the combination of exposure, sensitivity to PPPs and its distribution among species, as well as the resilience of the communities present, which in turn depends on the physiological state of the organisms in relation to other sources of stress associated with the pressure of PPPs. The impacts of PPPs on biodiversity are thus largely context-dependent and difficult to generalise. The pressure exerted by exposure to PPPs is combined in the field with other sources of stress, the two major being habitat destruction linked to agricultural intensification and urbanisation, and stresses linked to climate change. At a more local scale, the cumulative set of pressures results in changes in the balance of biotic interactions, which in turn can further aggravate the initial effects of PPPs.

## Simplified landscapes

In both agricultural and non-agricultural areas, numerous studies have shown the major influence of the composition and spatial arrangement of semi-natural habitats in landscapes on the abundance and diversity of communities and their ability to recover from an impact, as well as on associated ecosystem functions and services. As a habitat and food source for organisms, landscape characteristics play a key role in the organisms' ability to cope with the pressure of PPP exposure. A loss of refuge areas, their discontinuity, the lack of diversity within field margins as well as the lack of cover diversity within crops, tend to aggravate the impact of PPPs on biodiversity, as illustrated in Figure 19 for the example of terrestrial invertebrates. Such interactions are observed in the agricultural context, especially for bees. In the case of non-agricultural areas, the negative effect of

insecticide use in gardens on butterfly and bumblebee abundances is mainly seen in highly urbanised areas, where the surrounding habitats are less favourable to pollinators and do not, for example, allow recolonisation to occur. Modelling has shown that the presence of riparian woodland within landscapes can also limit the degradation of the ecological status of small rivers in some European countries, including France (Schriever and Liess, 2007).

**Figure 19. Landscape characteristics as factors influencing the effects of PPPs**



The simultaneous variation among the different characteristics of agroecosystems affecting biodiversity makes it difficult to identify and quantify the specific effect of each factor in isolation, and landscape variables are often considered as confounding factors when studies aim to assess the impact of PPPs on biodiversity. Most research therefore studies these aspects separately or tries to control one (or some) of these factors (e.g. experimental or semi-experimental treatments) and/or try to disentangle the factors (e.g. statistical analyses and meta-analyses, choice of sites with orthogonal characteristics) in order to try to rank their roles. For example, in a large European study, Geiger et al (2010) sought to disentangle the impact of different components of agricultural intensification (e.g. loss of landscape features, increasing plot size, use of fertilisers and PPPs) on biodiversity and biological control potential. Landscape characteristics, such as average plot size and percentage of arable land within 500 m, had a significant effect on species richness (especially plants and carabids) and aphid predation. Of the 13 intensification components measured, the use of herbicides, insecticides and fungicides had a consistent negative effect on biodiversity (wild plants, ground-dwelling carabids and birds). Insecticides also reduced the potential for biological control of pests.

However, a major underlying problem often overlooked in the literature is that landscape characteristics and PPP use intensities are correlated, with the most intensive uses occurring in simplified landscapes as a result of past and current intensification of practices. Within real world agroecosystems, organisms are thus subject to the joint pressures of landscape factors and the use of PPPs.

Several meta-analyses converge to show that the beneficial role of organic farming on biodiversity varies according to the surrounding landscape characteristics: the beneficial effect of organic farming on biodiversity is stronger when the percentage of the landscape occupied by arable plots increases, i.e. in intensive farming regions. This is less the case in complex and/or heterogeneous landscapes with small cultivated plots, including semi-natural habitats and with connectivity between habitats.

Modelling conducted on different taxa (e.g. birds, voles, hares) and different types of PPPs (e.g. insecticide, fungicide) also show that the intensity of PPP effects vary according to landscape composition characteristics (type of crops and proportions of cultivated areas/optimal habitats) and its spatial configuration (spatial arrangement between treated and untreated areas, connectivity/fragmentation). These studies underline the need to consider landscape factors (composition, configuration, connectivity) in population-level risk assessment, given their importance in influencing exposure and effects.

### Climate change

Sources of stress that may be related to climate change (e.g. increased average temperatures and temperature fluctuations, increased intensity of precipitation and droughts, flooding events, ocean acidification) appear most often in studies as causing increased sensitivity of organisms and vulnerability of populations to environmental toxins (including PPPs). Similarly, these substances may reduce the ability of organisms to cope with the consequences of climate change. For example, 83% of studies combining temperature increase and PPP exposure showed a synergistic interaction of these factors (Holmstrup *et al.*, 2010; Köhler and Triebkorn, 2013). Furthermore, sublethal effects of neurotoxic substances can induce a reduction in locomotor abilities and therefore the ability to move their home range, as has been shown in the damselfly (*Coenagrion scitulum*) exposed at the larval stage to the pyrethroid insecticide esfenvalerate. This may therefore lead to increased vulnerability to climate change for these populations.

In addition to the pressure it directly imposes on organisms, climate change is also expected to impact PPP use, and therefore the resulting exposure. It should also influence the fate of PPPs in the various environmental compartments. By modifying the physico-chemical and biotic properties of the latter as well as their dynamics (which can be observed, for example, with planktonic succession or the phenology of plants affected by climate change), it influences the kinetics and relative importance of the various transfer processes.

## Equilibrium shifts among biotic interactions

Vertical and horizontal biotic relationships both influence sensitivity to PPPs due to predation pressure or resource limitation. Thus, different levels of PPP effects have been observed, depending on parameters such as the availability of trophic resources, the presence/absence of predators, or other elements that determine the structure and dynamics of interactions between living organisms. Symbiotic interactions such as host-microbiota relationships, or host-pathogen interactions, can also influence or be influenced by PPP exposure.

Studies have been conducted on a case-by-case basis to observe the additive, synergistic or antagonistic nature of the relationship between the effects of certain PPPs and biotic interactions on certain species. For example, Oliveira dos Anjos *et al.* (2021) investigated the survival of daphnia exposed to a herbicide (diuron) and an insecticide (chlorpyrifos) in the presence or absence of a predator (notonectids). The findings indicate that the effects of the different stresses can be additive and synergistic in an environment where daphnia food resources are limited. Furthermore, Zhao *et al.* (2020) revealed that changes in the horizontal composition of a food web can increase or decrease the effects of PPPs. Expected consequences vary depending on whether the PPP induces mortality (a reduction in competition, compensatory effects at the population level, and access to the resource for a limited number of surviving individuals leading to a 'contest competition' situation), or a sublethal effect for all individuals (an increase in competition for the resource, leading to a 'scramble competition' situation). The interaction hypothesis between contaminant and density-dependence has also been proposed to explain the differences in impacts of the herbicide diquat on several life history traits of pond snails, depending on whether or not the experimental conditions favour intraspecific competition (Coutellec *et al.*, 2008).

Thus, in some cases, the weakening of some populations may benefit others within an ecosystem. For example, it has been observed that food limitation can increase individual vulnerability to PPPs in mayfly larvae (Hunn *et al.*, 2019), while conversely, in caddisfly *Limnephilus lunatus* larvae, the direct toxicity of the pyrethroid fenvalerate under chronic exposure conditions is compensated by the reduction in intraspecific competition resulting from mortality (Liess, 2002).

Although many studies show the effects of PPPs on biotic equilibria within communities, it is particularly difficult to form conclusions on the broader consequences for biodiversity. At this scale, field studies are necessary to identify the repercussions of the effects of PPPs on the entire range of interactions. For example, the link between the effects of PPPs and declines in terrestrial and aquatic invertebrates and birds has been established (see 'PPP's are a major contributor to the decline of certain taxonomic groups' section). The issue of what impact such a decline might be expected to have on the wider and longer-term dynamics of biotic interactions remains open.

## Reported improvements

This assessment of available knowledge reveals improvements at various levels. Bans on some of the most toxic substances generally result in a downward trend in the exposure of organisms to these substances. In addition, declines in use and improvements in biodiversity have been documented in some non-agricultural and agricultural systems, although no causal link has been established.

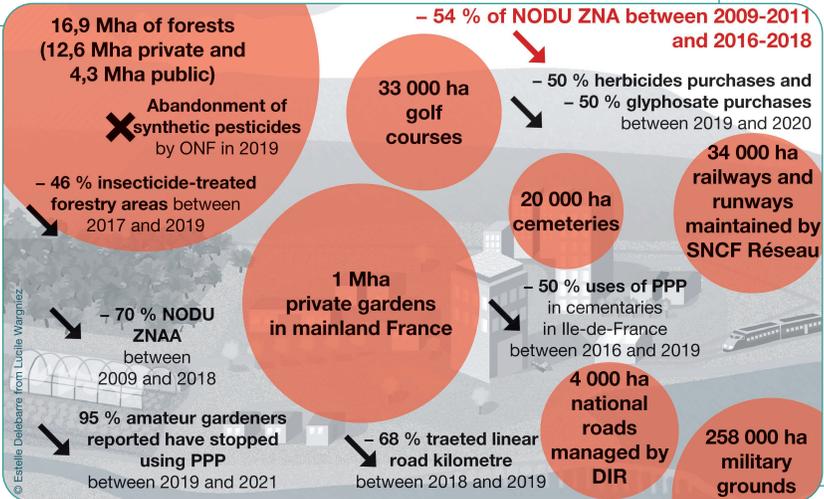
### ■ For certain substances

The banning of substances generally leads to a gradual reduction in contamination and its associated effects, without however leading to their complete disappearance, and with variable timeframes depending on the substances, the environments and the areas studied. For example, this disappearance may be slower in sediments or in certain organisms than in water. Thus, the trend towards the withdrawal of CMR Category 1 substances (carcinogenic, mutagenic and toxic for reproduction) has led to a decrease in the presence of these substances in surface aquatic environments over time. The 20% decrease in the IPCE (index of changes in pesticides in watercourses) between 2008 and 2017, monitored under the Ecophyto plan, reflects this outcome. This index, based on data from monitoring networks established under the WFD, depends directly on the methodology used, whether in terms of substances, sampling strategy or analysis capacity. These networks primarily monitor CMR substances, and weight the concentrations recorded according to their known toxicity. The withdrawal of active substances has thus led to an improvement in the index, in particular due to a downward trend in the concentrations of the most toxic and persistent herbicides, which are now banned. However, it is not yet possible to draw conclusions on the overall change in the degree of contamination of the environment, because these substances are being replaced by others, the behavior of which has not yet been observed in the environment or which are not monitored by the surveillance networks.

### ■ In non-agricultural areas

This state of affairs also varies according to the different types of non agricultural areas (reduction in use green spaces, but less so for road maintenance, for example). For this reason, non-agricultural areas could provide an opportunity to observe the effects on biodiversity, by comparing areas where PPPs have been withdrawn to those with continued use. The evidence obtained essentially shows two pathways towards the abandonment of PPPs. The first involves mechanical or thermal weeding, with an increase in the labour required. The second involves the use of biodiversity as a tool in its own right in the process of withdrawing PPPs, with selected plantings of species that prevent the spread of undesirable plants, and a review of the management guidelines for areas with greater tolerance for spontaneous vegetation. Thus, the impact of the PPP phase-out on biodiversity is the result of both the reduction of pressure caused by the substances and the implementation of management methods favourable to biodiversity.

**Figure 20. Geographical extent of non-agricultural areas and trends in the use of PPPs**



NODU: number of dose units; ZNA: non-agricultural zones; ZNAA: non-agricultural zones - non-professional use; DIR: interdepartmental road directorates; ONF: National Forest Office; SNCF: Société nationale des chemins de fer français (National society of French railroads).

Very few academic references deal with the impact of PPPs on biodiversity. The use of PPPs in non-agricultural areas is mainly dealt with in relation to human health concerns and rarely with biodiversity. Conversely, references dealing with biodiversity in non-agricultural areas most often deal with the organisation of these areas, their uses and knowledge of their biodiversity, and rarely document links to PPP application. This question has therefore been addressed mainly from non-academic sources that allow changes in PPP use to be documented in parallel with that of biodiversity in non-agricultural areas.

## Improvements made, and persistent difficulties on the scientific front

### Collection of data on PPP use and contamination

Since the beginning of the 2000s, networks for monitoring the state of the environment have gradually placed more and more emphasis on PPPs. The number of substances monitored in each environmental compartment has increased, even though research into transformation products remains very patchy. With the development of analytical techniques using chromatography (liquid or gas) coupled with mass spectrometry, increasingly

long lists of PPPs from different chemical families are being monitored in continental and marine aquatic environments. Although air is not subject to monitoring under any regulatory framework, campaigns undertaken since the early 2000s by *Associations agréées de surveillance de la qualité de l'air* (Approved air quality monitoring associations; AASQA) provide one of the most extensive databases on the presence of PPPs in Europe, although it lacks uniformity in the methodologies used. The establishment of a standardised permanent monitoring system was agreed to at the national level in 2021. With regard to soils, which are also not subject to compulsory monitoring of PPPs, monitoring campaigns were initiated in the early 2000s by the Soil Quality Measurement Network (RMQS), but these only involved a limited number of PPPs and transformation products that are now banned, namely organochlorine insecticides, triazine and phenylurea herbicides and copper. The measurements led to the mapping of concentrations, in particular for copper and lindane. Based on the RMQS, *the Phytosol project* extended this monitoring to include 110 different currently authorised PPPs (selected on the basis of recommendations from Anses). The first sampling was conducted in 2019 and 2020 at 50 sites within this network, mainly in field crops and viticulture; the results of these analyses were not yet available at the time of writing.

However, the list of compounds to be monitored must be regularly updated in order to reflect the current usage of PPPs. This is achieved by various means within the framework of the Ecophyto plan. The BNVD, the main reference for the overall monitoring of quantities of PPPs sold, has been updated since 2009. It is based on declarations used to calculate the diffuse pollution tax applied to PPPs, and the information collected has been gradually refined. In 2012, usage data became spatially referenced based on the purchaser's postal code. This data has become standardized and has, since 2015, been usable. PPPs are also the subject of statistical surveys on cultivation practices carried out by the Ministry of Agriculture. Based on sampling, they allow average trends to be established at national and regional levels on the number of annual treatments and the quantities used. Finally, a unified methodology for calculating the TFI (treatment frequency index) has been established to provide a baseline and facilitate the observation of changes or comparisons of PPP use within a group of farms or a type of crop.

## ■ Basic knowledge of community and ecosystem ecology

The importance of indirect effects, combined with the difficulty of explaining and anticipating their dynamics and consequences on the functioning of ecosystems, is an obstacle to reaching a quantifiable conclusion on the specific role and relative share of PPP impacts on biodiversity and ecosystem functions, a point that is repeatedly raised in the literature. This difficulty relates, among other things, to the lack of fundamental knowledge on the functional role of species at different ecological levels (population, community, ecosystem, biome), and to the lack of analysis of the interrelationships between them and the biotope, in both aquatic and terrestrial natural environments.

This is illustrated in the case of invertebrates, which account for most animal biodiversity and play an essential role in the functioning of ecosystems. Although the effects of PPPs are well documented for these organisms, the concepts of regional diversity (gamma diversity) or dissimilarity between communities (beta diversity) are never addressed. Moreover, despite the existence of different indicators of functional diversity (e.g. Schmera *et al.*, 2017) for aquatic macroinvertebrates, there remains a need for further development of descriptors capable of translating the activities of organisms in the ecosystem (behaviour) in order to study and quantify their contribution to ecological processes such as bioturbation, degradation of organic matter, and pollination, which contribute to different ecosystem functions. Furthermore, the often-promised development of explanatory approaches such as AOPs is characterized by a profound lack of knowledge of the mechanisms that cause unintended effects at the sub-individual and individual levels (depending on the metabolisms of each species), and of the consequences for individual fitness. This results in an inability to predict the impacts on population and ecosystem parameters. In addition, AOPs do not address the problem of assessing indirect effects.

## I Standardisation and usability of tools

The literature review has shown that scientific research on the contamination of environments by PPPs and its impacts on biodiversity and ecosystem functions and services has led to conceptual and methodological developments and innovations. However, most of the methods developed and used in the scientific sphere are not standardised, which sometimes limits the scope of the results they generate, particularly in the context of operational approaches including regulatory approaches.

This observation is particularly well illustrated by the case of 'omics' methods, which rely on technologies that are constantly being developed, both in terms of amplification and sequencing techniques and in terms of bioinformatics approaches and tools for sequence analysis. As a result, most of them are not yet mature enough to be standardised. They also lack a reference framework for translating the results of the various 'omics' methods into biological and ecological consequences. It would therefore be appropriate to establish a guide to good practice (or even to move towards standardisation) in the acquisition and processing of data generated by these methods, in order to facilitate their use and apply them routinely to a wide range of organisms. The example of the application of metabarcoding to benthic diatoms for the estimation of the ecological quality of rivers (Vasselon *et al.*, 2017) illustrates the promising nature of this type of approach. For this type of organism, and more generally for primary producers, the parallel development of imaging tools could also enable progress in taxonomic recognition, phenotyping of higher plants and broader observation at the scale of biofilms.

Similarly, the PICT method, which has been successfully used to provide *in situ* information on the impact of PPPs on natural microbial communities exposed in various ecosystems (Bérard *et al.*, 2021), is not yet used (or usable) in the regulatory environment,

mainly due to the lack of standardised protocols or a reference system for interpretation. These aspects are more advanced for the SPEAR approach, which has been widely applied to aquatic macroinvertebrates at different scales, including at the French national level. Its applicability to benthic diatoms, for which it has only very recently been applied in Australia (Wood *et al.*, 2019), has yet to be demonstrated in the French (or more generally European) context.

## I Modelling potential and data accessibility

Modelling is repeatedly mentioned as a solution for integrating the processes involved at different spatial and temporal scales, and in particular for linking the dynamics of use, exposure and effects.

Modelling has the potential to assess the effectiveness of devices aimed at limiting PPP transfers (e.g. drift control equipment, untreated areas, hedges, wetlands, ditches) and to parameterise their characteristics. However, at the catchment scale, the translation of models into operational tools remains a challenge. The lack of data as well as the estimation of uncertainties in results remain obstacles to the use of these models for prioritising different changes in practices and layouts at this scale. Furthermore, the coupling of different types of models is often invoked. For example, it is proposed to couple hydrological, atmospheric, and also agronomic transfer models (Voltz *et al.*, 2019).

Six main categories of models exist to assess the ecotoxicological and ecological effects of PPPs, and are summarised in Table 3 (Larras *et al.*, 2022a).

Among these models, spatial population models have potential either by incorporating landscape characteristics (Topping and Weyman, 2018) or by modelling spatial dynamics from individual data.

At the landscape scale, this includes estimating the contamination of organisms, the toxicity of a PPP, or demographic effects by taking into account the variability of landscape patterns and exposure (Topping *et al.*, 2015). At this scale, modelling approaches are particularly relevant to better understand the different factors involved in the decline of certain species and to prioritise their impacts.

However, modelling currently does not allow transgenerational effects to be integrated, and community and food web models are not sufficiently developed at relevant scales to simulate ecological processes, particularly in a multi-stress context.

Furthermore, improvements in the assessment of PPP impacts requires the coupling of ecotoxicological models (which capture the effects of PPPs) with ecological models (which provide information on the interactions between organisms and the functions they provide).

Bioeconomic models have also been suggested as a means of broadly assessing the impacts of PPPs on ecosystem services. However, such linkages still face the challenge of formalising impact indicators and shared interpretation frameworks.

**Table 3. Main categories of ecotoxicological and ecological models identified in the literature**

Category	Model	Characteristics	Relevant outputs
QSAR	Quantitative structure-activity relationship	Relationship between chemical structures and activities of PPPs	Acute toxicity, mutagenic properties, bioconcentration factors
DR and TKTD	Dose-response (DR)	Relationship between exposure concentration and individual response or effect, after a fixed exposure time	Survival, growth, reproduction, mobility, enzyme activities, feeding rate etc.
	Toxicokinetic-toxicodynamic (TKTD)	Relationship between exposure and individual effect, including the time course of exposure and effects	Survival, growth, reproduction, mobility, enzyme activities, feeding rate etc.
Population	Population	Relationship between individual effects and demographic response, including ecological conditions for population occurrence	Population growth rate, population density, risk of extinction, demographic recovery time, changes in population structure (including spatial distribution)
Multi-species	Species sensitivity distribution (SSD)	Effects at the species assemblage level (without consideration of species interactions)	Probabilistic assessment of a hazardous concentration for a certain % of species in the assemblage
	Food webs (or food chains)	Ecological interactions between species: 'simple' (e.g. predator-prey relationship) or 'complex' (network of ecological interactions, with inclusion of abiotic factors) models	Biomagnification and indirect effects via trophic cascades
	Community model		Direct effects on species and/or indirect effects on relationships between species, at the community level and/or for ecosystem services
Landscape	Habitat models at local, regional or national scales	Ecological impacts at the landscape scale, incorporating the spatial dimension (implicitly or explicitly)	Demographic responses within different habitats, maintenance of non-target species, contamination levels
Mixture models	Concentration addition (CA), independent action (IA), TKTD	Effects of PPP mixtures on individual traits	Synergy, antagonism, neutrality

It is important to emphasise that while modelling offers major potential for the production and use of knowledge of the impacts of PPPs, this potential can only be achieved with the support of basic knowledge of the processes involved, and empirical data that is essential for the development, parameterisation and validation of the models.

Finally, the translation of these tools to types of treatment (e.g. biocontrol), or to contexts that differ from those in which they were developed (e.g. the French overseas territories), remains an important issue. Approaches by environment type (with associated models, scenarios and data sets) would allow the prioritisation of PPP use contexts on which to focus efforts.

## 6. Interactions between science and regulation

PPPs are developed at the interface between contradictory requirements: they must be effective on the targeted organisms, but without unacceptable effects on others. The regulatory framework for products is thus based on criteria for identifying unintended effects and establishing their degree of acceptability. In this area, the independent health authorities<sup>23</sup> play a central role at the scientific level (by funding studies and publishing articles) and at the regulatory level (by establishing regulations that are binding on stakeholders and assessing the validity of applications submitted on the basis of these regulations). Three main levels structure this interaction between science and regulation for the assessment of the impacts of PPPs:

- the framing of the assessment method and the studies required. On the basis of the existing literature, the health authorities establish the requirements to be met by petitioners regarding the transfer processes and the types of effects on biodiversity to be taken into consideration. These requirements in turn generate scientific activity into knowledge of the processes and the corresponding effects;
- the determination of effect thresholds. Standardised tests or measurements are used to standardise the protocols for the detection of contamination and its effects, as well as their interpretation in terms of acceptability/inacceptability using thresholds;
- the final decision on authorisation. It takes into account, in connection with the above elements, other areas of concern related to PPP use (political, economic or legal). Thus, exemptions may be granted at the national level for substances not approved at the European level (e.g. neonicotinoids), or conversely, restrictions may be imposed at the national level for substances that have been approved at the EU level (e.g. sulfoxaflor, glyphosate).

Thus, while the risk assessment is based on ecotoxicological grounds, the decision to authorise is the responsibility of the risk manager, who integrates other economic and social parameters. The final decision is therefore the result of a compromise between objectives that may sometimes be contradictory.

These processes lead to two-way interactions between science and regulation, which are fraught with issues, and have been the subject of research at the interface of the humanities and social sciences and ecotoxicology. The literature on these subjects converges toward criticism of the current regulations. The inadequacies of the system have attracted much more attention than its improvements or successes. This criticism of the assessment of PPP effects on biodiversity is also largely based on the more general criticism of the effects

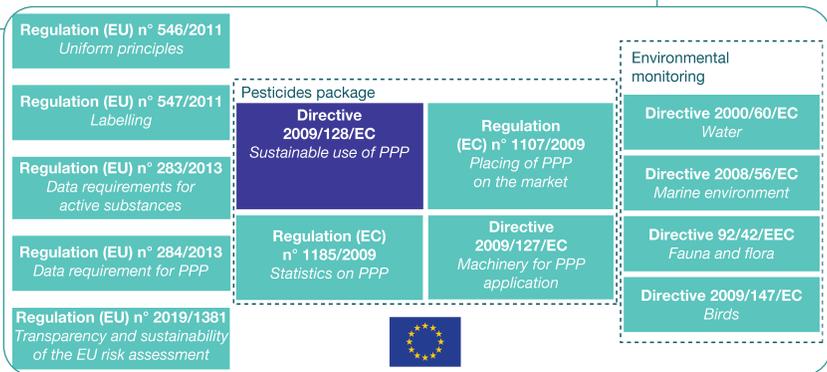
23. Anses at French level, EFSA at European Union level

on human health. This work highlights the gap between the ambition of strong protection enshrined in the principles of the regulations and the impacts resulting from the use of PPPs. They highlight the tendency for regulatory complexity to increase as a result of these two-way interactions. They also analyse the constraints that result in the exclusion of some available scientific knowledge in the academic field from the decision-making process. Furthermore, although assessments of observed impacts on the environment are gradually being improved in the regulatory domain, they remain difficult to incorporate into the decision-making process. On the basis of these observations, many studies suggest avenues for improvement, whether in terms of scientific assessment methods or in terms of the decision-making processes that lead to the approval of uses.

### Requirements and complexity of PPP regulations

PPP are subject to a complex system of regulations, as shown in Figure 21, the main element of which stems from the Pesticides Package at the European level. This set of regulations covers Directive 2009/128/EC establishing a “framework for Community action to achieve the sustainable use of pesticides”, Regulation (EC) No 1107/2009 (2009b), which sets out the rules concerning the placing of PPPs on the market, and a series of regulatory texts relating in particular to application machinery, PPP labelling and statistical monitoring. They also include more cross-cutting rules, such as regulations on the establishment of risk assessment bodies and processes, and in particular the activities of EFSA and Anses. As chemical contaminants, PPPs are also subject to monitoring requirements within the regulations dealing with the environment and biodiversity protection, which stem from the European framework directives on water, marine environments, habitat protection, and the protection of flora, fauna and birds.

**Figure 21. Main EU legislative texts on PPPs and biodiversity**



The European regulations governing the marketing of PPPs set high standards for the protection of human health and the environment. Article 4 of Regulation (EC) No 1107/2009 (2009b) stipulates that substances must have "no harmful effect on human or animal health and no unacceptable effects on the environment" when used in accordance with "good plant protection practices". This regulation is thus recognised in the literature as providing a high degree of protection for biodiversity compared to most other major jurisdictions, and the EU has withdrawn (and continues to withdraw) many substances that are problematic in terms of human or environmental health, but which are still permitted in, for example, the USA, Brazil or China (e.g. acetochlor, atrazine, clothianidin, thiamethoxam, imidacloprid).

### Specific regulations relating to biocontrol

The French regulations applying to biocontrol products (Article L.253-6 of the CRPM) are specific and seek to facilitate their release onto the market (LAAAF law No. 2014-1170). They benefit from a reduced tax for applications for approval and authorisation, a reduced assessment period and various exemptions (Article R.253-11 of the CRPM). They are exempt from the ban (Article L.253-51 of the CRPM) on discounts, rebates and refunds, and special sales conditions applied to other PPPs. Approval is not required for use as a service provision when the product does not carry any hazard warning (Article L.254-1 of the CRPM). Some advertising, which is prohibited for PPPs, is authorised (Article D.253-43-2 of the CRPM) for biocontrol. The use of these products is also exempted from the obligation to implement measures to protect people in the vicinity of inhabited areas or areas used for recreational purposes (article L.253-8 II of the CRPM). Biocontrol PPPs can be sold and used by public entities, and used in green spaces, forests, roads or public footpaths (article L.253-7 of the CRPM). They are also exempted from actions aimed at reducing the use of PPPs and from PPP saving certificates (CEPP, articles L.254-10 to L254-10-9 of the CRPM). Once approved, biocontrol PPPs are listed in Annex II of Regulation (EC) No 889/2008.

Microorganisms, semiochemicals and natural substances are covered by Regulation (EC) No 1107/2009 (2009b) and are subject to a list updated monthly at the national level and disseminated via a note from the Ministry of Agriculture and Food. Non-indigenous macroorganisms that may present specific risks to the environment (e.g. invasive species) are not subject to the same regulations. Since 2012, they have been subject to Decree n° 2012-140 of 30 January 2012 on the conditions for authorising the entry into the French territory and the introduction into the environment of non-indigenous macroorganisms useful to plants, particularly in the context of biological control. However, those which have been introduced for several years, before the date of entry into force of the decree, and which do not present any particular risk, are exempted from an application for authorisation for entry or for introduction.

However, the sophistication of European rules has led to the development, over the last twenty years, of a regulatory armoury whose complexity sometimes hinders its intelligibility. For example, the tendency to withdraw the most problematic substances is counterbalanced by the proliferation of exemption schemes. Similarly, the methods set out in the guidance documents produced by EFSA or Anses are not always followed through, as some of these documents are not adopted by the risk management authorities (e.g. the 2013 EFSA guidelines on pollinators). The intelligibility of the assessments is also affected by the large amounts and complexity of data provided in the application and MA (Marketing Authorisation) documentation, which are therefore difficult to verify (Robinson *et al.*, 2020).

Despite ambitious regulatory targets, PPPs have been shown to play a role in reducing biodiversity and degrading some ecosystem functions (see section on 'Impacts on ecosystem functions'). Consequently, there is a discrepancy between the ambitions set out in EU law and the environmental degradation caused by PPPs.

### Available scientific knowledge not being considered

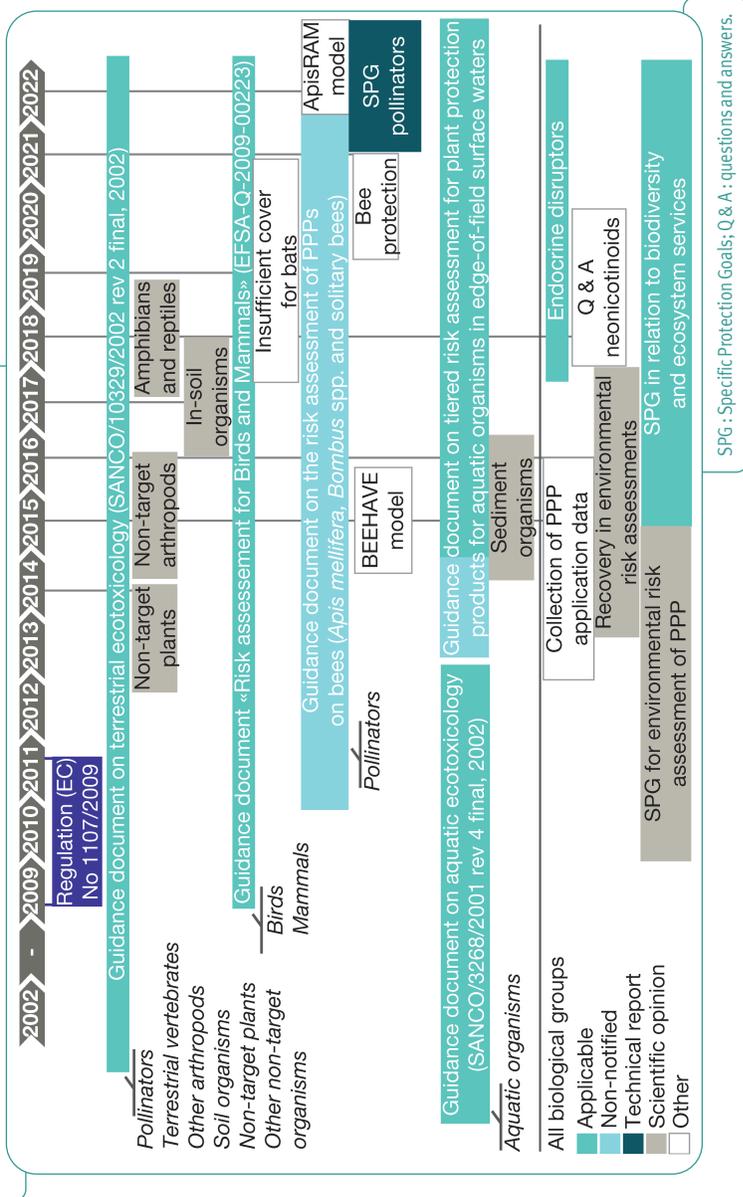
The scientific principles applied to the regulatory framework fail to include much of the knowledge available in the academic sphere, whether because of the decision-making processes, the time frame required to integrate the available knowledge into these processes, or the constraints specific to the regulatory framework (*a priori* evaluation by substance, with no consideration of the context of use). The analysis of this type of phenomenon has recently been formalised in the field of agnotology, or the study of ignorance, as a process of creating ignorance (Jouzel, 2019).

### Resulting from decision-making processes

The standards that govern the requirements for conducting the tests and ecotoxicological studies that comprise risk assessments are largely based on Organisation for Economic Co-operation and Development (OECD) standards, which play a role in harmonising standards at the global level to facilitate trade. However, the literature reveals a lack of transparency and management of conflicts of interest in the development of these standards. This has important consequences for the types of effects and risks considered, and therefore for the results of assessments. Certain fields of knowledge thus appear to be excluded from risk assessments when they have not been developed according to regulatory standards (Jouzel, 2019).

The difficulties posed by what is still considered to be an overly vague management of the links of interest within the panels of experts who establish standards and carry out risk assessment are thus a subject widely addressed in the literature. The interactions between assessment, industry, the market and the State are described as leading to what Demortain and Boullier (2019) call 'assessment by the market'. The assessment of

Figure 22. Evolution of the main scientific requirements for risk assessment under the EU regulatory framework (adapted from Lanras *et al.*, 2022a)



products and their risks is carried out in an environment that is constructed at the confluence of scientific competence, administrative problems and the market.

The principle of the petitioner being responsible for proving the safety of the substance for which approval is sought is considered by some authors to introduce a conflict of interest bias. These authors have drawn on various studies to show that studies conducted or funded by PPP developers are more likely to conclude that the substance is safe than studies conducted by scientists independent of the industry.

However, changes have more recently been introduced in relation to all of these procedural issues through the revision in 2019, following the 2017 Glyphosate Citizens' Initiative, of the Regulation governing the evaluation process at the European level (Regulation (EU) No 2019/1381, 2019). These amendments aim to address the aforementioned issues, in particular by strengthening transparency and access to information, the possibility of conducting studies independently of the petitioners, the procedures for recruiting experts and for managing links of interest.

### **I Due to the time frame in which standards and rules are developed**

The integration of scientific knowledge into the regulatory process is a long process that introduces a time lag between the availability of knowledge and methods and their integration (Dedieu, 2021). This time lag includes the stages of gathering and appraising the research, and if necessary standardising the systems for producing and interpreting the results, transcribing them into peer-reviewed guidance documents, and then submitting them for adoption by the EU's political bodies when required by the regulations. Thus, more than ten years can routinely pass between the release of knowledge and its inclusion, in practice, in the risk assessment. Figure 22 shows the gradual build-up of scientific resources that underpin an assessment process within the EU regulatory system, and the timescale over which this occurs. Furthermore, some guidance documents must undergo an adoption procedure by the committee representing the EU Member States before they are made mandatory. In some cases, this step can result in an additional delay (e.g. risk assessment for aquatic organisms at the edge of surface waters) or even in the rejection of the proposed guidelines in the case of the 2013 document on risk assessment for pollinators. In this case, it led to a revision of EFSA's approach, and an extension of its work on pollinators for another decade.

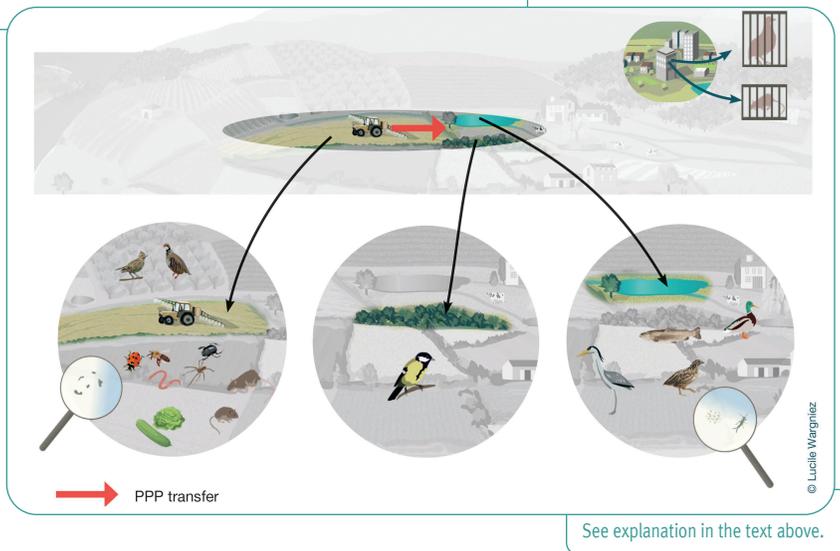
### **I Due to specific constraints within the regulatory frameworks**

#### **Standardization of the tools used**

Standardisation of tests is required to ensure the validity of results. However, this standardisation involves constraints on the types of effects and the types of organisms tested (adaptability to confinement, length of the life cycle, time to observe the effect). A compromise is therefore sought between the representativeness of the phenomena studied and their observability by repeatable experimental designs. Thus, the ecotoxicological report is

based on tests carried out on species that are presumed to represent the diversity of species in the field and their place in the ecosystem. Figure 23 illustrates this focus of environmental risk assessment within the cultivated area and transfers in the immediate vicinity, as well as the selective nature of the species studied in the cultivated area, in its immediate surroundings and in adjacent wetlands. Finally, the toxicity of the products is only assessed on model species adapted to laboratory conditions, as illustrated in Figure 23 by the cages located in the buildings. Some studies carried out in 'cosms' using modelling tools aim to extend the analysis to more complex scales, but these are only implemented where the first tier approach has shown unacceptable effects (see 'Tiered approach' section). In particular, landscape and trophic interactions, multiple uses in space and time are not taken into account.

**Figure 23. Scope of the *a priori* risk assessment**



Finally, the interpretation of the test results is based on the determination of protection goals, the level of which is not subject to consensus (e.g. level of effect considered to be unacceptable, proportion of the observed population or community on which this level of effect is reached).

### Limited use of modelling

Modelling offers the possibility of completing the assessment by drawing, in particular, on knowledge acquired experimentally. However, models are sometimes based on assumptions that may not be valid in nature, and on physiological, biological and ecological data that are still very incomplete. In addition, a large proportion of existing models are not used for risk assessment. As part of this CSA, a comparison was made between the range of models

referenced in the literature and those actually used in the regulatory environment. The latter are essentially limited to QSAR (quantitative structure-activity relationship) models, DR (dose-response) models and SSD (species sensitivity distribution) models. More complex models such as TKTD (toxicokinetic-toxicodynamic), 'population', 'community', 'food web', 'mixture', and landscape scale models as well as exposome models are only rarely used (Larras *et al.*, 2022b). Although this gap has been identified for more than a decade, the acceptance and validation of these complex models by risk assessment agencies remains limited. There is insufficient information and expertise available to attest to the validity of these tools in a regulatory framework. The complexity of the models is cited as a barrier to their use, with the required skills sometimes restricted to the small group of teams involved in their development, themselves nationals of a small proportion of EU Member States.

### Assumptions and uncertainties

At the time of the pre-market environmental risk assessment of a PPP, the only precisely known elements are the active substance, its physicochemical characteristics (which partly determine its fate in the environment), its ecotoxicity as assessed on the basis of the standardised tests mentioned above, and the main formulations in which it is likely to be employed. The remainder of the assessment is necessarily based on assumptions that cover the manner and conditions of application, the characteristics of the recipient environment, the potentially exposed species and their exposure pathways, other substances with which the product may interact, etc. The assessment also does not take into account the effects of the product on the environment. Furthermore, the assessment does not account for the chronological sequence of PPP applications that may limit the possibility of recovery, which leads to an underestimation of exposure. For these reasons, risk assessments incorporate safety factors, which consist of applying a multiplier to the results obtained.

These numerous uncertainties can lead to a discrepancy between the assessed impacts and the reality observed in the field. An insufficient or total lack of disclosure of the scientific uncertainty resulting from these assumptions for any risk assessment is identified in the literature as likely to undermine trust in the assessment agencies.

### Tiered approach

In the first instance, the exposure scenarios considered are so-called 'worst-case' scenarios, based on strong exposure assumptions, but carried out on a limited number of species and criteria (notably only direct effects). The results of these trials are accompanied by safety factors to take into account the difference in sensitivity between individuals and species. They are sometimes considered to be overprotective due to the lack of realism in the exposure and sensitivity levels considered but, conversely, they are sometimes considered to be insufficiently protective because they do not consider the diversity of the species present and their interactions.

This approach can be refined by additional tests at different levels called 'Tiers' (Tier 2 for additional laboratory tests, Tier 3 for tests in microcosms, mesocosms or in the field).

These tests aim to better define the reality of the exposure, but focusing on even fewer species. In the most complex cases, studies under semi-natural conditions or in the field can be produced, but no standardised protocol or precise recommendations are currently available for this. Furthermore, these additional assessment levels are only required in cases where the first test step does not yield results that would allow approval. In cases where the first stage of testing produces effect levels below the acceptable thresholds, no further study is carried out on real-life exposure contexts or species interactions. In fact, an examination of the applications for approval or renewal of 318 substances evaluated by EFSA between 2011 and 2021 shows that less than half of the evaluations are based on field or ‘cosm’ trials that would allow effects to be observed at community level. In all cases, the analysis of indirect effects (e.g. alteration of food resources) and more generally of trophic interactions in an ecosystem remains very limited, although it is explicitly mentioned in Regulation (EC) No 1107/2009 (2009b).

### Excluded scientific approaches

These standardised assessment procedures cannot be transposed to higher biological scales, nor to ecosystem functions and services, and do not allow for the complexity of the effects of PPPs on biodiversity. This explains why the conclusions established on the basis of regulatory tests are sometimes contradicted by field observations documented elsewhere in the literature (on the basis of field trials or post-authorisation monitoring in the case of renewal dossiers), which are often based on different protocols from those used for regulatory purposes (e.g. species, developmental stages tested, test conditions). The following approaches are thus identified as being difficult to reconcile at this stage with the requirements for generating scientific knowledge within the regulatory framework:

- knowledge based on field observations, which can only be used for *a priori* evaluation in the case of approval applications for renewal, or for authorisations of new uses or new formulations for an already approved active substance;
- results based on methods specific to the research project from which they originate, but which have not been standardised;
- results that establish correlations, but which do not demonstrate the mechanism of impact of the evaluated PPP on the observed biodiversity;
- systemic approaches that consider interacting factors but do not isolate the specific contribution of the evaluated PPP to the observed effects;
- modelling tools developed at scales that integrate population, community or landscape dynamics with a more complex approach to exposure, but which require skills for their use that are often restricted to the research teams that developed them.

Lastly, in addition to the studies specifically developed by the applicant for marketing authorisation, the academic literature must be considered in the procedure, in particular to guide the search for suspected potential effects based on observations made on substances or uses with similar characteristics, etc. However, it is hampered by the difficulty of systematically and reproducibly selecting the most relevant studies from a non-standard

set of scientific outputs, which address potential real-world effects in a heterogeneous manner. This literature is therefore only marginally exploited in practice. It should also be emphasised that no literature is available for new substances.

### **Disconnection between pre-market and post-market assessment**

The literature describes citizens' movements, which increasingly draw on available scientific results, but which have not been taken into account in the regulatory process, in order to alert public authorities to the need to strengthen the control of certain substances with regard to their undesirable effects. At the same time, initiatives have begun to make this post-authorisation observation more systematic and to take better account of warning signals. This is the purpose of phytopharmacovigilance (PPV) and its linkage with, on the one hand, the monitoring networks set up under the regulations on biodiversity protection and, on the other hand, the authorisation processes. However, this approach encounters the limits of existing monitoring networks and the fact that these results are still only partially considered for risk assessment when applying for marketing authorisation renewal.

### **Limitations of environmental monitoring**

For many years, the scientific community has highlighted the lack of monitoring in the field, at the landscape level and over the long term, of the fate of PPPs in soils, air, freshwater and marine aquatic environments and biota, as well as their effects on organisms and communities. This monitoring is largely based on the obligations arising from the European directives on the environment, on fauna and on flora: WFD, MSFD, Habitats Directive, and Birds Directive.

To systematically describe the impacts of PPPs, monitoring systems are based around the selection of substances, species and contexts considered to be the most at risk, and on the criteria and indicators considered to be the most revealing (e.g. environmental quality standards, biodiversity indicators). This selection is the result of a compromise between the representativeness of the impacts as they occur in the environment, on the one hand, and the reliability of the measurements, the ease of interpretation of the results, the possibility of establishing temporal trends and comparisons between sites, on the other. These compromises give rise to shortcomings that are widely documented in the analysed literature. In particular, the choice of substances monitored still neglects, despite some progress, the diversity of substances currently in use as well as transformation products. Monitoring is still largely focused on substances already banned, but which remain a priority in terms of their toxicity.

The species and communities most commonly identified as poorly monitored are heterotrophic and autotrophic microorganisms, particularly symbiotic microorganisms, protozoa,

zooplankton, wild pollinators, aquatic vertebrates, amphibians, reptiles and bats (Mougin *et al.*, 2018).

In general, many gaps in fundamental knowledge of ecosystems and their functioning, which are essential in order to better direct environmental monitoring efforts, have been highlighted. For this reason, systems based on a broader approach to field observation, such as *Zones Ateliers* (Workshop Areas) and participatory research programmes, are often singled out for their ability to better capture the diversity of effects and their dynamics.

## ■ Limits to the inclusion of monitoring in risk assessments

Although mentioned in Regulation (EC) No 1107/2009 (2009b), monitoring for PPP effects after they have been released onto the market has not been the subject of any specific recommendation at the EU level. At the French national level, the PPV was set up in 2014 and implemented by Anses since 2015. This system is unique in Europe in its objective of centralising and cross-analysing data on documented adverse effects for each PPP, produced by different partner networks. These data mainly come from the monitoring networks mentioned above (see section 'Limitations of environmental monitoring'), but also from the funding of additional studies and research programmes, a reporting portal for any organisation or citizen who has observed undesirable effects, as well as a bibliographic monitoring system to harness published data or identify areas for improvement. This data is assembled to establish a phytopharmacovigilance sheet for each substance, which is posted on the Anses website.

However, the information currently used only covers a limited portion of the total information available, due to constraints relating to the degree of scientific validation of the reliability of the data in question, and the priority given to parameters for which temporal trends can be established and for which reference thresholds exist. Therefore, PPV essentially covers information relating to environmental contamination, and remains very limited with regard to the effects on biodiversity. At the same time, much of the existing data remains underused (e.g. Ozcar, Critical Zone Observatories - Applications and Research network; *Zones Ateliers*; the SAGIR network; and monitoring conducted by the MNHN, the French National Museum of Natural History). Looking to the future, work is underway to progressively improve the use of this available knowledge, such as that gathered by the MNHN as part of its participatory research programmes on biodiversity monitoring (e.g. STOC, Vigie-Nature) cross-referenced with spatially-referenced data on PPP sales (BNVD). Studies of this type, combining monitoring of organisms over time as well as PPP applications and measurements of PPP residues, can lead to clearer inferences between exposure and effects in the field.

Following this same objective, monitoring of unintended effects of PPPs on farmland biodiversity is being implemented as part of the Écophyto plan, with a focus on several non-target taxonomic groups (earthworms, field edge plants, beetles and birds) on 500 farm plots (Andrade *et al.*, 2021). The objectives are to detect changes in the frequency

or abundance of indicator species along with simultaneous changes in agricultural practices, including PPP applications, and to improve knowledge. The first results after four years of study show a higher species richness in organic farming than in conventional farming, mainly related to the higher number of species at the field edges. This open-air laboratory has helped in identifying the key elements for carrying out this type of study. Problems were nevertheless identified in relation to missing explanatory variables and the varying identification skills of observers for certain taxa, although the data relating to the agricultural context were sound and consistent.

In principle, the regulations stipulate that the identification of undesirable effects may lead to measures to restrict the use of PPPs to limit risks and protect ecosystems. At this stage, although the information sheets drawn up under the PPV for various substances are sent to the risk assessors when an application for re-authorisation is examined, they are still too limited to provide a genuine complement to the identification of impacts on biodiversity.

### Most frequently identified avenues for improvement

Faced with the inadequacies of the impact assessment processes used in the regulatory framework, these proposed improvements have been developed by experts in ecotoxicology and the humanities and social sciences. They can be classified by differentiating between recommendations concerning the knowledge incorporated into the regulatory framework and those concerning changes to the framework itself.

#### ■ In the knowledge domain

##### Types of knowledge to be considered

Some authors believe that the inadequacy of the current assessment procedures is due to the fact that the only scientific data included is from the life sciences, without considering social data (Hamlyn, 2017), which is part of the notion of sustainable development invoked by Directive 2009/128/EC (European Commission, 2009a). Hamlyn (2017) thus advocates for a holistic approach, which includes economic and social data in the approval process. This type of approach is presented as being able to better integrate cost-benefit considerations into the decision-making process.

In addition, many authors from the sociology, law, political science and geography fields have highlighted the lack of inclusion of the socio-ecological complexity inherent in the effects of PPPs on biodiversity and associated ecosystem functions and services. Systems thinking, as observed for example in the field of beekeeping (Suryanarayanan, 2013), is mostly ignored in the regulatory procedures. The epistemic form of knowledge observed among beekeepers identified as 'naturalists' (Adam *et al.*, 2020), namely the set of concepts, measurements and interpretations specific to their social group, relies on knowledge

based on field observation, which considers the actual contexts of the observation of natural dynamics. This so-called 'integrated knowledge' can lead to research on the longer-term (i.e. more than a few weeks) effects of certain PPPs, on cumulative effects and on sublethal effects. Similarly, non-academic knowledge (e.g. farmers, citizens' groups, residents, NGOs), especially knowledge based on experience, is not considered.

### Scientific tools that can be mobilised

Given the particular constraints of the regulatory framework discussed above, not all of the knowledge available in the academic domain can be directly used. The adaptation and generalisation of models and methods based on a systemic approach (e.g. multiple, chronic, ubiquitous, multidimensional environmental exposures) are made difficult by the lack of large-scale observation data. Moreover, there are no simple criteria for assessing and quantifying the effects of PPPs on biodiversity or on ecosystem functions and services.

The literature does, however, point to possible ways to improve the scientific tools used in the regulatory environment. These have been discussed in the methodological sections of this CSA and are summarised in Table 4. Most of the avenues for improvement identified in the literature are also compiled in the recent work of Topping et al (2020).

Numerous avenues are thus available for the development of tools, both methodological and technical.

The use of new methodologies within microcosm and mesocosm experiments, as well as studies based on life traits, has the potential to integrate more functional responses and to better address biotic interactions and the indirect effects of PPPs. The transgenerational effects approach can be applied to organisms whose genome is known, in order to assess the resilience and recovery capacities of exposed populations. The protocols for experimental tests could thus be adapted, for example, by the choice of biological and physiological traits of the species under study, the duration and repetition of exposures, and the exposure routes, in order to achieve an assessment that better reflects the ecological reality.

In the longer term, AOP-type approaches, which are essentially focused on the individual, could help to investigate the biological causalities and empirical evidence that support (or refute) the relationships between responses measured at different levels of biological organisation, thus better linking experimental data and field observations. AOPs are based on various molecular initiating events: oxidative stress, serotonin transporter inhibition, DNA damage, mitochondrial dysfunction, endocrine mechanisms (e.g. inhibition of chitin synthase or  $11\beta$ -hydroxysteroid dehydrogenase, activation of the juvenile hormone receptor) and epigenetic mechanisms (inhibition of DNA methyltransferase). The approach can be applied to fish, gastropods and bivalves among the molluscs, freshwater microcrustaceans, higher plants and microalgae. The development of these AOPs on species already used as ecotoxicological models increases the benefit of the approach by increasing the knowledge base for these models. Overall, the study of metabolic pathways using *in vitro* methods (cell cultures) or metabolomics approaches (at the individual level)

**Table 4. Scientific tools that can be used within the regulatory processes for approvals and marketing authorisation (MA) and for monitoring (PPV). Specific examples of tools are indicated in brackets**

Scientific tools that can be mobilised	Multiple stresses	Consideration of mixtures and exposure history	Functional consequences
Mesocosms <sup>a</sup>	MA	MA	MA
Screening using species with a known genome ( <i>in vitro</i> hormone receptor tests) or bioinformatics			
'Omics' tools (all 'omics', functional and structural)	MA	MA	MA
Biomarkers (AChE)	MA	PPV	MA
Functional trait approaches (diagnostic tool)	MA		PPV
Trophic dynamics studies			
Multi-residue, non-targeted monitoring, effects monitoring (PICT, SPEAR), field data	PPV	PPV	
Modelling	MA (CA, IA)	MA (CA, IA)	

would enable the behaviour of PPPs in organisms to be assessed *a priori*, and the toxicity of the transformation products formed and interspecific differences to be understood.

In terms of modelling, the assessment of environmental exposure is based on models from the 'FOCUS' group which, although they have the advantage of simulating PPP transfers and the dynamics of associated exposure history at the end of a treated plot, do not however make it possible to describe all of the processes involved in the fate and transfer of PPPs (e.g. particle transport), nor the great diversity of existing agricultural practices. Furthermore, no model integrates the land-sea continuum. The principal models used to assess the effects of PPPs are QSAR, DR, TKTD and SSD (Larras *et al.*, 2022b). So-called 'population', 'community' and 'landscape' models are still not widely

Parameters to develop					
Complex levels of organisation	Choice of species, and life stages included	Adaptive, transgenerational effects	Indirect effects	Sub-lethal effects	Acceptable concentrations
MA	MA PPV	MA	MA	MA	MA
	MA			MA	
MA PPV	MA PPV	MA PPV		MA	
				MA PPV	
MA	MA	MA	MA (trophic aspects)	MA PPV	
MA	MA PPV		MA PPV		
PPV	PPV			PPV	PPV
MA (ALMaSS)	MA PPV				MA (TKTD, DEBtox)

ALMaSS: Animal Landscape and Man Simulation System; CA: concentration addition; DEBtox: Dynamic Energy Budget; IA: independent action; TKTD: toxicokinetic-toxicodynamic.

<sup>a</sup> Mesocosm studies are in some cases required by Tier 3 regulations to assess the ecotoxicity of PPPs (see section 'Tiered approaches').

used at the regulatory level, despite their acknowledged value (Topping and Luttik, 2017; Topping *et al.*, 2020; Larras *et al.*, 2022). Spatially explicit population models represent a major avenue of development for testing various hypotheses on the basis of theoretical scenarios, but also for assessing risk for terrestrial vertebrates, amphibians, reptiles or chiropterans (Ockleford *et al.*, 2018; Hernandez-Jerez *et al.*, 2019). At the landscape scale, some authors recommend that future risk assessments use multiple scenarios representative of a wide range of crop and landscape conditions to avoid the occurrence of locally unacceptable risks.

In addition to the methodological and technical innovations that can be deployed, the importance of the strategy that should guide the implementation of investigation tools is

widely emphasised. Multi-residue analyses, non-targeted analyses and analyses based on effects are thus highlighted for the development of biomonitoring that can be interoperable on a large scale, over the long term, and among different trophic levels. The challenge is to better detect situations or molecules at risk, or changes in contaminant exposure dynamics, provide information on multiple exposures and monitor the effectiveness of management measures. Regarding the choice of species tested, various recent publications propose, based on the work of Dietzen *et al.* (2014) and regulatory requirements, relevant focal species that are better targeted for cereal crops (granivorous birds such as the grey partridge) by integrating agricultural practices (e.g. presence before or after sowing) (Bonneris *et al.*, 2019).

Risk assessment would also benefit from studies using species that are more representative (of the ecosystems studied and of the assumed exposure), studies focusing on the adaptation of organisms to PPPs through transgenerational effects (organisms with known genomes, laboratory model animals for screening), studies on the resilience/recovery of exposed populations, and studies on the direct and indirect effects of PPPs. Studies based on life traits and functional approaches should also be integrated, as well as the study of interspecific, interguild and intraguild interactions (which are particularly important in biocontrol). There is also a need to study the effects of PPPs at the food web level.

In general, to take better advantage of field data, which is inevitably more heterogeneous than standardised laboratory tests, a framework for producing results and managing, sharing and processing data needs to be developed that is better suited to more systemic problems. These could take the form of guidelines, at an intermediate level between no standardisation and strict standardisation (in particular for the use of modelling methods for impact assessment, 'omics', and environmental DNA), which would improve the comparability and compilation of results, while allowing the operational methods to be adapted to the context. An example of recent progress in this direction is the Pepper platform (Public-private platform for the pre-validation of endocrine disruptors characterization methods), launched in 2019.

Finally, the development of studies relating to the evaluation of ecosystem services would enable the analysis of impacts to be made more comprehensive by highlighting their consequences on some of the issues specifically identified by stakeholders, as proposed by EFSA (Benford *et al.*, 2016). However, this analytical framework remains difficult to make systematic for approvals and marketing authorisations, given the still very limited consideration of the ecological processes on which it is based, and the difficulties of balancing competing services that benefit different stakeholders.

## **I In the regulatory domain**

Many authors point to the need to increase the transparency and independence of the conduct of assessments, including the accessibility of the data used in applications. However, most of the articles on this subject predate the implementation of Regulation (EU) No 2019/1381 (2019), which is intended to address these concerns. Known as the

'Transparency Regulation', it aims to strengthen the transparency and sustainability of risk assessment within the food production chain<sup>24</sup> at the European Union level, and came into force on 27 March 2021. Robinson et al. (2020) also advocate using panels of scientists who are independent of the economic interests of the petitioners to look for biases, invalid or outdated assumptions and possible violations of the precautionary principle in the methodologies used, in order to review them independently of the administrative authorities. The same type of proposal was recently presented in a notice issued by the *Commission nationale de déontologie et des alertes en santé publique* (National Commission on Ethics and Public Health Alerts), in relation to public confidence in the evaluation process for the renewal of the glyphosate approval<sup>25</sup>.

Various proposals concern the manner in which risk assessment studies are conducted, which to date have been carried out by the applicant for marketing authorisation. In order to better safeguard the independence of such studies, it is proposed that they be paid for by industry, but commissioned by the assessment agencies and conducted in independent public laboratories. The applicant would no longer be able to choose the laboratory or scientists responsible for these studies, nor the design and conduct of the studies or the interpretation of the results.

Various studies have revealed the roles played by coalitions of stakeholders (researchers, beekeepers, NGOs, politicians advocating environmental action, companies) in the production and use of research in order to intervene in the regulatory process. Some case studies, particularly those on the authorisation processes for neonicotinoids and glyphosate, demonstrate how stakeholders such as professional federations, environmental associations and consumer groups can intervene from outside the regulatory evaluation process and influence decision-making. For example, in the case of neonicotinoids, analyses have focused on the discrepancy between the decisions taken by managers and the results of the *a priori* evaluation. This work describes how, in response to concerns expressed by stakeholders in favour of protecting pollinating insects (Demortain, 2021), some of these substances were banned in France in the 2000s and 2010s, even though they had been approved at the European level. The concerns raised led to the gradual withdrawal of various substances within the neonicotinoid family at the European level in the 2010s. However, in 2021 and 2022, in response to concerns regarding the protection of sugar beet crops, temporary exemptions were granted at the national level under Article 53 of Regulation (EC) No 1107/2009 (2009b) (so-called '120-day' exemptions). In France, these are accompanied by provisions that do not allow their renewal beyond three years, and include a targeted research programme. These conflicts over neonicotinoids also led EFSA to publish updated guidelines for the risk assessment of PPPs for bees in 2013, but this scientific framework has not been adopted by the EU Member

24. <https://www.anses.fr/fr/content/le-r%C3%A8glement-europ%C3%A9en-sur-la-transparence-un-nouveau-cadre-pour-l%E2%80%99%C3%A9valuation-des-risques-et> (accessed 9/01/2023).

25. <https://www.alerte-sante-environnement-deontologie.fr/deontologie-et-alertes-en-sante-publique-et-environnement/travaux/avis-rendus/article/avis-sur-les-conditions-de-la-confiance-des-citoyens-vis-a-vis-du-processus-d> (accessed 9/01/2023).

States at the regulatory level. A revision of EFSA's approach was therefore required. These findings have led various authors to advocate opening up the assessment process to a wider range of participants and knowledge than has so far been the case under the current arrangements (Mohring *et al.*, 2020).

### Specific regulations relating to non-agricultural areas

The European framework directive on the sustainable use of pesticides, adopted in 2009, provides for the implementation of measures to protect sensitive groups of people from PPPs. In France, the regulations concerning the use of PPPs in agricultural and forestry products have been gradually evolving for more than ten years towards the abandonment of the use of these products. The decree of 27 June 2011, also known as the 'Public Places Decree' (ALP), initiated restrictions on use in areas open to the public. This concerns places usually frequented by vulnerable people and places open to the public, where certain products are prohibited (e.g. schools, recreation areas, surroundings of buildings for the elderly, sick or disabled).

Law no. 2014-110 of 6 February 2014, known as the 'Labbé Law', set two major deadlines for limiting the use of PPPs in non-agricultural areas, with the exception of biocontrol products, products that can be used in organic farming, and low-risk products. The dates of entry into force of these provisions were brought forward by Article 68 of Law No. 2015-992 of 17 August 2015 on Energy Transition for Green Growth (LTE). Thus, the over-the-counter sale to individuals and the use of these products by public persons were prohibited from 1 January 2017. The sale, use and possession of these same products were banned for private individuals as of 1 January 2019. However, a series of exceptions were maintained (e.g. private areas such as the surroundings of business premises or condominiums, cemeteries) but these are now included in the decree of 15 January 2021 which extended the list of areas concerned. On 1 July 2022, as set out in the decree, all public and private places frequented by the public or used collectively were banned from the use of PPPs, with the exception of playing fields (e.g. football, rugby, hockey), lawn tennis courts, racecourses and golf courses. For the latter, the ban will apply from 1 January 2025. After this date, the use of synthetic PPPs will only be permitted for roads that are inaccessible to the public and for which there is a safety issue associated with vegetation control (e.g. railway verges and airport runways) and for the uses listed by the Ministries of Sport and the Environment for top-level sports fields, when there is no alternative technical solution that would provide the quality required for official competitions.

# Conclusions

## PPP contamination is known to affect all environments

Since the early 2000s, the monitoring of PPP contamination of the environment has been progressively strengthened, by expanding the list of substances monitored and the different matrices sampled. Improvements in analytical techniques and the development of multi-residue approaches, as well as improvements in sampling strategies, for example through the use of passive samplers, have also led to improvements in detection and quantification, including for chronic contamination at low concentrations.

Current knowledge reveals proven contamination of all environments (terrestrial, aquatic, atmospheric) by various PPPs. Accordingly, numerous active substances and some of their transformation products have been found, particularly in the soil and small streams and headwater rivers in areas dominated by agriculture, but also in downstream waters, sediments and marine environments (particularly coastal waters). In addition, trophic transfer of some of these substances has been proven. This transfer contributes to the spread of the contamination within the food webs of these various environments, up to the large predators. This contamination of biota confirms the exposure of a wide range of organisms to PPPs, even at a distance from the application sites, through to fish of the deep ocean. Airborne pathways also contribute to the dispersion and redeposition of compounds. A significant number of substances are found in the atmosphere, including in rainwater, at distances that can vary from the immediate edge of the plot to the regional or even continental scale, depending on the compounds.

This contamination usually takes the form of complex mixtures involving several active substances (including substances that are no longer approved for use but persist in the environment), co-formulants, adjuvants and transformation products. However, the latter are still largely unknown and under-sought, as are adjuvants and co-formulants.

Current environmental monitoring strategies, however, remain inadequate, particularly regarding the exposure of organisms to the most recently introduced products, which include biocontrol solutions. Simultaneous monitoring of substances in all matrices, including biota, is not currently performed. This would give a better picture of the dynamics of contamination. Methods such as passive samplers and multi-residue analyses without *a priori* choice of targeted substances (including, among others, transformation products) are now available but not yet deployed on a large enough scale to enable monitoring to better reflect reality. More widespread implementation of this type of approach will also raise the issue of managing and sharing the data generated, as is the case for 'omics'

methods. Transfer models can also contribute to the choice of substances to be monitored and to setting the spatial and temporal strategy to be implemented.

### **The state of knowledge in the French overseas territories remains highly incomplete**

Although monitoring networks provide information on the contamination of aquatic environments in the French overseas departments, scientific studies on environmental PPP contamination in the overseas territories are rare. Most of the identified studies concern contamination in Martinique and Guadeloupe by chlordecone, with particular attention given to contamination of biota. The specific characteristics of the various French overseas territories reflect the characteristics of their agricultural activities, except in the uninhabited territories located in the sub-Antarctic zone, which are contaminated by organochlorine PPPs linked to the long-distance transport of these molecules.

Despite proven contamination, to our knowledge no study has documented its effects on biodiversity in the natural environment of the French overseas territories. However, environmental (and particularly climatic) conditions and the types of crops in these territories differ from those in mainland France. These differences limit the transferability of knowledge and methods produced in mainland France. Indeed, studies of PPP effects on biodiversity require the use of new biological models that accurately represent the biodiversity of the overseas territories. To do this, substantial investment is required to acquire knowledge on the ecology and physiology of the species that comprise this biodiversity, in order to allow their use as model species in ecology and ecotoxicology. This effort is necessary both for knowledge acquisition and for risk assessment purposes because, even if knowledge acquired in territories that are similar in terms of geography, climate and/or practices can provide some insight, the different situations justify the development of specific knowledge. The geographical distance from France also hampers the development of this research, but this can be addressed by setting up incentives to encourage collaboration between local research teams and others in mainland France. This was the case, for example, with the calls for projects under the successive national chlordecone plans.

### **PPPs contribute to the weakening of biodiversity**

PPPs negatively impact biodiversity, through both direct and indirect effects, with the importance of indirect effects increasingly being highlighted. In the agricultural areas of mainland France, robust results show the involvement of PPPs in the decline of populations of terrestrial and aquatic invertebrates and birds. Although this decline dynamic has been observed for several decades, studies over the last fifteen years have documented

in a convergent manner the direct and indirect processes that link the use of PPPs to the weakening of these populations. Numerous studies have thus enabled the identification of exposure pathways that had not previously been considered, of sublethal effects disrupting the endocrine, nervous or immune systems of organisms or their interactions with microbiota, which had previously been ignored, or of indirect effects through the reduction of trophic resources or alteration of habitats, which had been underestimated. The same types of processes have begun to be highlighted for amphibian and bat populations in agricultural areas, although for these species the results have yet to be consolidated due to the less extensive scientific literature.

Such effects have implications for the functioning of ecosystems and the ecosystem services they provide, which are relevant to a broad spectrum of species due to the interactions that underpin ecological functions. The selectivity of substances' mechanisms, i.e. their ability to target a narrow spectrum of species, is thus often contradicted in the longer term by observable unintended effects that had not been identified *a priori*. This re-evaluation of selectivity has sometimes been linked to evidence of sublethal effects at the organism level on the endocrine, nervous or immune systems, or even at the holobiont level in terms of interactions with microbiota. However, these types of sublethal effects are still very poorly studied for a large proportion of biological groups. Selectivity also appears to be undermined by the indirect nature of the effects suffered by non-target organisms, due to their interactions with the target organisms. However, the ecosystem dimension of the vast majority of studies aimed at assessing the impacts of PPPs on biodiversity and the functioning of contaminated environments is still too limited to elucidate and anticipate the dynamics of the indirect impacts, which are nevertheless often predominant.

The relative contribution of PPPs to the erosion of biodiversity is thus difficult to establish as a whole, given the multifactorial context combining different toxic (PPP and other contaminants) as well as physico-chemical and biological pressures (e.g. climatic constraints, modification of habitats, pathogens, invasive species, organic inputs).

The extent of the geographical areas affected by the use of PPPs and the repeated nature of the applications limit the possibilities of mitigating the effects through the resilience of the non-target populations affected. The chemical pressure induced by the use of these substances is also correlated with other important pressure factors, such as the simplification of landscapes leading to the disappearance of habitats of ecological importance. The influence of the landscape has been emphasised in many studies, and the association between the use of PPPs and the absence of interconnected refuge areas has been highlighted as a particularly damaging combination. Moreover, in combination with other more widespread sources of stress such as climate change and the emergence of pathogens, PPPs contribute to an increase in the vulnerability of some ecosystems, particularly agroecosystems. These pressures together also induce adaptation processes in certain species, which alter community equilibria. The use of PPPs can also lead to a deterioration in crop health when it provides a selective or competitive advantage to pest species that are less sensitive or less exposed because of their biological characteristics.

For example, heavier and less mobile species of terrestrial invertebrates may be more exposed during treatments. Given that lower mobility as a biological trait is more prevalent in predators and detritivores (including a large proportion of beneficial organisms) than in phytophagous organisms (including a large proportion of pests), an advantage is conferred on the latter. However, studies analysing these types of response dynamics to PPP pressure under natural conditions are still very limited.

### **PPPs reduce the capacity to provide ecosystem services**

While the literature reveals that PPPs impact some ecosystem functions, the resulting consequences for ecosystem services are still only addressed for a limited range of services, with a focus on crop production, pollination and biological control. This knowledge reveals a tension between crop production and other biodiversity services. Indeed, PPPs replace the ecosystem service of pest regulation in the short term and allow the elimination of targeted organisms to increase agricultural production. In doing so, however, they undermine ecosystem regulation, with negative consequences for agricultural production. They also affect the provision of other services such as pollination, the formation and maintenance of soil quality, and certain cultural services such as water quality (recreational fishing and bathing) and landscape amenities. Thus, the message from the reviewed literature is that the preservation of ecosystem services requires a reduction in the pressure exerted by PPPs.

In terms of risk assessment, a disparity has been noted between the expectations for an ecosystem services approach to assessment that integrates and prioritises different issues, and the scientific work available on the specific impacts of substances or families of PPP substances on ecosystem services. Indeed, existing work in this area is limited to a few specific links between a family of substances and a service, for example the impacts of neonicotinoids on pollination. Many obstacles stand in the way of *a priori* assessment of the consequences of introducing a PPP onto the market on ecosystem services as a whole. These obstacles particularly concern the conceptual and operational barriers between disciplines dealing with ecosystem services and those dealing with ecotoxicology, but also the lack of tools for prioritising potentially competing ecosystem services.

### **Impacts are highly dependent on the methods and context of use**

The impacts observed vary according to the substances and their characteristics, but also according to the methods and agro-pedoclimatic contexts of PPP use. These sources of variability relate in particular to the timing of PPP applications in a given location, the methods of application and the characteristics of the environment in which they are applied. These parameters influence product losses, dispersion dynamics in the environment and the level of exposure of non-target organisms together with their degree of sensitivity and

vulnerability. The extent of the resulting ecosystem-level effects depends on the interactions between the biodiversity components present. Although these mechanisms cannot be described exhaustively, the available knowledge highlights some situations that most often harm biodiversity, including applications carried out under unfavourable meteorological and agro-pedoclimatic conditions, repetitive applications that do not allow the non-target species to recover, and the lack of possible refuge within and outside the plot (simplified landscapes with no continuity of refuge areas).

Although available data on crop protection practices has been considerably improved since 2005, it is still insufficiently precise with regard to the temporality and geographical location of treatments to analyse the influence of the context on the fate, transfer and effects of substances in real contexts.

## Instruments to partially mitigate the impacts

### I Choice of substances and biocontrol

The *a priori* assessment of the ecotoxicity of substances establishes fundamental benchmarks to guide the choice of substances with a view to limiting undesirable impacts. This choice must, however, be considered in the light of a set of parameters that are not known *a priori*, such as application methods, the extent and repetition of use, and the agro-pedoclimatic and landscape characteristics of the environment to which the substance is applied. It is therefore crucial to consider the changes in crop protection practices by taking into account all of the methods deployed, and not just the substitution of one substance by another. This simple substitution may in fact lead to the displacement of effects, as shown in some studies, instead of the mitigation initially intended.

Biocontrol is promoted as an alternative capable of mitigating the unwanted effects of PPPs. In this domain, the literature mainly focuses on the development of solutions, and documents their modes of action and efficacy according to different usage methods, or even in terms of their interactions with other biocontrol agents. Very little research has looked at the consequences of these treatments on the environment, except in the case of the organisms that have been used for the longest time (e.g. *Bacillus thuringiensis*, *Harmonia axyridis*). However, the use of living organisms (microorganisms and macroorganisms) in biocontrol adds a unique dimension compared to conventional PPPs, as they can multiply, move and colonise other environments. Regarding natural substances, the limited results to date indicate that while most of them have low ecotoxicity, others (e.g. abamectin or spinosad) have a toxicity equivalent to or higher than that of their synthetic counterparts.

### I Limiting transfers

Research has intensified over the last twenty years in order to better understand the transfer of PPPs and the associated means of mitigation, particularly on the basis of *in situ*

experimentation. The spatio-temporal dynamics of the fate of PPPs in the environment are also the subject of modelling that makes it possible to link different scales and processes. Different measures (application methods, soil management, planning, and remediation) to limit PPP transfers are being tested in the field, but none of them totally eliminates them. Their effectiveness depends in particular on the combination of several measures (complementary and not additive) and on their geographical positioning at the catchment scale.

## **I Landscape characteristics**

Landscape characteristics are cited in many studies as a major factor influencing the impacts of PPPs on biodiversity, aggravating it in the case of simplified landscapes, while mitigating it in the case of landscape mosaics with multiple interfaces between treated and untreated areas and connectivity with refuge areas. The landscape thus plays a role both in direct effects, by limiting the exposure of organisms through interception of the chemicals, and in indirect effects, by preserving food resources and habitats. This influence is highlighted by modelling that links the dynamics of contamination and effects, by integrating a classification of landscape characteristics to assess the regulating influence. However, such approaches are still *ad hoc*, and require the development of large-scale field observation systems.

### **In non-agricultural areas, a redesign of management methods**

In non-agricultural areas, the trend towards a reduction in PPP use was initiated by certain local authorities or territorial bodies in the mid-2010s, and has continued since then, particularly with the restriction on use introduced by the Labbé law from 2017 and gradually extended to most types of non-agricultural areas. Very little scientific work has been conducted on biodiversity changes in relation to this reduction in use. The results of studies collected from outside the academic field show little use of biocontrol products and agents to replace PPPs that are now banned, and a reduction in the need for them based on three main approaches: greater acceptance of spontaneous vegetation, an increase in the use of labour and skills, and the use of biodiversity as a tool for managing areas, by planting species chosen in accordance with the desired functions of the area. These findings show the potential of biodiversity as an instrument for managing ecosystem functionality.

### **The ambitious objectives of the PPP regulatory framework have not been fully met**

The risk assessment processes prior to the release of new PPPs form part of the regulatory system at the European and French national levels, which affirm the need to protect human health and the environment. Consequently, this regulatory system is recognised

as one of the most stringent in the world. However, academic studies have shown that many effects that threaten the preservation of biodiversity are not foreseen in the assessment process prior to the release of PPPs on the market, which does not allow the complexity of real environments to be integrated. Due to the small number of variables and species studied, it is therefore after the PPP has been put into use that deleterious environmental effects are often observed. However, the monitoring of unintended effects after the release of PPPs onto the market has to date centred mainly on the results of the monitoring of environmental contamination and on the effects on human health. Available knowledge on the impacts on biodiversity is therefore only marginally considered when applications for the renewal of approvals are examined.

### The use of existing knowledge for regulatory purposes needs to be better organised

The body of work analysed reveals that much available knowledge is not used for *a priori* assessment of the risks associated with the use of PPPs. In particular, the choice of species to be tested could be revised to better represent not only their taxonomic diversity, but also their functional diversity. The experimental test protocols could also be adapted in terms of the biological and physiological traits of the species tested, the duration and repetition of exposure, and the exposure routes, thus obtaining an assessment that better represents ecological reality. Sublethal effects are not adequately assessed, especially with regard to disturbances (e.g. behaviour, immunity, physiological condition), which may affect the fitness of species and their role in ecosystem functioning. At the landscape scale, some authors recommend that future risk assessments should use multiple scenarios representative of a wide variety of agricultural practices and pedoclimatic contexts. In terms of post-approval surveillance, improvements in knowledge have enabled the development of innovative methods that are already or could be applied to improve the biomonitoring of exposure to and effects of PPPs in ecosystems. For example, the implementation of calibrated *in situ* bioassays (e.g. the Afnor AChE standard for gammarids) can resolve the interpretation difficulties posed by the influence of confounding factors (other pollution and environmental stresses) in field monitoring. Bioassays applied to the study of the decomposition of organic matter in soils are also the subject of international ISO standards. The PICT method offers a framework for observing effects on the structure and diversity of communities, particularly for microorganisms. The same applies to the SPEAR method, which is mainly applied to aquatic invertebrates but which could be extended to other organisms (e.g. diatoms). The mobilisation of 'omics' approaches provides a complementary means of understanding the specific ecotoxicological responses to the pressure exerted by PPPs. Finally, the integration of tools based on biological effects, including approaches that take into account responses at a functional level, would make it possible to complete the methodologies for monitoring environments, not only for their chemical status, but also for their ecological status.

However, the mobilisation of these approaches in regulatory processes requires collective organisation to establish implementation protocols and shared interpretation frameworks. To this end, international standards are usually sought (e.g. OECD or ISO), although their limitations in terms of addressing the complexity of effects on biodiversity have been highlighted. The mobilisation of scientific communities is essential in order to recommend test protocols that are more relevant to the effects on biodiversity. Furthermore, intermediate degrees of harmonisation could be developed, such as the pre-validation of methods. This has recently appeared in the regulatory landscape, and is similar to that proposed for endocrine disruptors by the Pepper platform.

Finally, the humanities and social science literature analysed highlights the growing role of coalitions of stakeholders (environmental and consumer associations, NGOs, companies) in the process of mobilising scientific knowledge in the regulatory field. The mobilisations of these actors, particularly around the regulation of neonicotinoids and glyphosate, illustrate the role played by these stakeholders in the use of data produced by the scientific community, as well as the growing importance of concerns that lead to the production and recognition of new knowledge. Some authors promote a broadening of the types of stakeholders and knowledge to be considered when determining the regulatory status of substances. Thus, consultation could be envisaged in order to define precise scientific objectives combined with the mobilisation and pooling of resources around these objectives, as well as dedicated experiments, to enable different scientific communities to combine their specific insights.

### **Taking better account of the complexity of exposures and effects**

The analysis of research conducted over the last two decades shows that there are still significant knowledge gaps, whether in terms of types of PPPs (biocontrol), transformation products, types of organisms (e.g. amphibians, reptiles, less studied symbiotic organisms such as corals, mycorrhizae, lichens, microbiota), types of environments and regions (e.g. marine, French overseas territories) or types of effects (e.g. sublethal, synergistic, cumulative). Scientific approaches are addressing increasingly diverse levels of organisation and interaction, but the proliferation of studies has translated, overall, into considerable heterogeneity at this point. It is therefore necessary to promote more integrated research strategies to enable the complex reality of PPP exposure and effects to be taken into account. Sets of indicators should be combined to integrate the direct ecotoxicity of substances and their indirect effects, depending on the characteristics of the system in question (e.g. landscape, agroecosystem). To this end, studies based on different climatic scenarios, different land use scenarios, and the spatial heterogeneity of contamination or effects could be developed.

Research on the impacts of PPPs on biodiversity appears to be highly compartmentalised, particularly in terms of the types of organisms studied, or the types of environments or

environmental matrices taken into consideration. The assessment of the effects of PPPs on the various components of biodiversity is made less relevant by the absence of a more integrated perspective based on the complexity, vulnerability and sustainability of ecosystems. The scope of this assessment is also constrained by the difficulty of addressing issues related to biotic and abiotic environmental continua, which are crucial in the context of biodiversity conservation. In order to better address these dimensions, it is necessary to adapt the range of scientific disciplines involved to better understand the diversity of the observed effects. For example, the study of the impacts of PPP use on certain ecosystem functions, such as the regulation of water flows in soils and sediments, albedo (reflection) or the formation and maintenance of soil and sediment structure, requires the involvement of disciplines such as physics and hydrophysics, which have rarely, if ever, been involved in ecotoxicology studies.

Assessing the effects of PPPs on biodiversity and ecosystem functions and services therefore requires the pooling of resources from different disciplines around shared objectives. Certain research networks, such as the French RECOTOX initiative, which could be extended to the marine environment, represent a first step in this direction. However, it would be beneficial to use instrumented and/or long-term monitoring study sites, such as those connected to the LTER (Long Term Ecological Research Network) or within certain *Zones Ateliers* (Workshop Areas), which are suited to the study of PPP contamination and its impacts. These mechanisms could be strengthened and extended by means of dedicated incentives. From this perspective, the reviewed literature clearly reveals an insufficient exchange and sharing of tools and concepts specific to each of the disciplinary fields that deal with the same issues. The study of the impacts of anthropogenic pressures requires interdisciplinary approaches and the pooling of knowledge on the functioning of living organisms, hydro-biogeochemical functioning, social functioning, associated economic issues, associated legal concepts, etc., in order to better document the implications of policy actions.

### Linking the study of agricultural systems to that of ecosystems

This CSA has focused on the impacts of PPPs on biodiversity and ecosystem services, which assumes a causal chain restricted to one factor (PPP use) and one direction (from PPPs to biodiversity). However, the use of PPPs cannot be dissociated from other parameters within agricultural production systems, which also influence the status of biodiversity and the functioning of ecosystems. In turn, biodiversity changes affect the conditions of agricultural production and crop health, which are themselves linked to the issue of food systems on the one hand, and available factors of production on the other.

These interrelated dynamics require the involvement of other scientific communities and the use of other knowledge. The results of this CSA on the impacts of PPPs should

therefore be considered in relation to those of other recent studies, particularly in the context of the Ecophyto plan's calls for projects, such as the CSA 'Protect crops by increasing plant diversity in agricultural areas'<sup>26</sup>, or the 'Growing and Protecting Crops Differently' priority research programme<sup>27</sup>, as well as the foresight study 'Pesticide-free European Agriculture by 2050'<sup>28</sup>.

Finally, from a 'One Health' or even 'Eco Health' perspective that addresses ecosystem functions, human health and animal health in an integrated manner, links involving shared exposure dynamics or common types of effects at the sub-organism level have yet to be studied.

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26. <https://www.inrae.fr/actualites/augmenter-diversite-vegetale-espaces-agricoles-protoger-cultures>

27. [https://www6.inrae.fr/cultiver-protoger-autrement\\_eng/Programme](https://www6.inrae.fr/cultiver-protoger-autrement_eng/Programme)

28. <https://www.inrae.fr/en/news/european-chemical-pesticide-free-agriculture-2050-results-groundbreaking-foresight-study>

# Acronyms and abbreviations

- Afnor** *Association française de normalisation* (French Standards Association)
- AMM** Autorisation de mise sur le marché (marketing authorisation)
- AMPA** Aminomethylphosphonic acid
- Anses** *Agence nationale de sécurité sanitaire de l'alimentation, de l'environnement et du travail* (French Agency for Food, Environmental and Occupational Health and Safety)
- AOP** Adverse outcome pathway
- AWBZ** Artificial wetland buffer zone
- BCF** Bioconcentration factor
- BMF** Biomagnification factor
- BNVD** *Banque nationale des ventes par les distributeurs agréés* (National Database of Sales by Authorised Distributors)
- BZ** Buffer zone
- CBD** Convention on Biological Diversity
- CICES** Common International Classification for Ecosystem Services
- CMR** Carcinogenic, mutagenic, or toxic for reproduction
- CNEP** *Campagne nationale exploratoire des pesticides* (national exploratory pesticide campaign)
- CRPM** *Code rural et de la pêche maritime* (Rural and Maritime Fishing Code)
- CSA** Collective scientific assessment
- DBZ** Dry buffer zone
- EC** European Commission
- EC<sub>50</sub>** Half maximal effective concentration
- Efese** *Évaluation française des écosystèmes et des services écosystémiques* (French assessment of ecosystems and ecosystem services)
- EFSA** European Food Safety Authority
- EU** European Union
- Ifremer** *Institut français de recherche pour l'exploitation de la mer* (National Institute for Ocean Science)
- INRAE** *Institut national de recherche pour l'agriculture, l'alimentation et l'environnement* (French National Research Institute for Agriculture, Food and Environment)
- IPBES** Intergovernmental Science-Policy Platform on Biodiversity and Ecosystem Services

**ISO** International Organization for Standardization

**ITS** Internal transcribed spacer

**IUCN** International Union for Conservation of Nature

**LOD** Limit of detection

**LOQ** Limit of quantification

**MNHN** *Muséum national d'histoire naturelle* (French National Museum of Natural History)

**MSFD** Marine Strategy Framework Directive

**NGO** Non-governmental organisation

**OECD** Organisation for Economic Co-operation and Development

**PICT** Pollution-induced community tolerance

**POP** Persistent organic pollutants

**PPDB** Pesticide Properties DataBase

**PPP** Plant protection product

**PPV** Phytopharmacovigilance

**RECOTOX** *Initiative en éco-toxicologie pour suivre, comprendre et réduire les impacts des pesticides dans les socio-agroécosystèmes* (Eco-toxicology initiative to monitor, understand and reduce the impacts of pesticides in socio-agroecosystems)

**RMQS** *Réseau de mesure de la qualité des sols* (Soil Quality Measurement Network)

**SDHI** Succinate dehydrogenase inhibitor

**SPEAR** Species at risk

**SSD** Species sensitivity distribution

**STOC** *Suivi temporel des oiseaux communs* (Temporal Monitoring of Common Birds)

**UN** United Nations

**WBZ** Wet buffer zone

**WFD** Water Framework Directive

**WoS** Web of Science™

# Glossary

Boldface type indicates terms defined elsewhere in this glossary.

## **Adjuvant**

A preparation or substance with no plant protection activity of its own that can be added extemporaneously or incorporated as a co-formulant into plant protection products to enhance their physico-chemical properties (e.g. oils, various surfactants). Adjuvants have been defined in Regulation (EC) No 1107/2009 (2009b) Article 2(3) as not being synergists or safeners, but rather preparations that facilitate the wettability or adhesion of PPPs, or prevent foaming. The release of adjuvants on the market is regulated.

## **Bioaccumulation**

A gradual increase in the amount of a substance in an organism or part of an organism that occurs because the rate of uptake exceeds the organism's ability to eliminate the PPP. Active ingredients and their **transformation products** can be more or less bioaccumulative in body tissues and along food webs. This accumulation is quantified via different factors: bioconcentration factor (BCF) for accumulation in organisms following their exposure in the environment (e.g. fish, earthworm); biota-sediment accumulation factor (BSAF); biomagnification factor (BMF) by food depending on the trophic level considered (increase in the concentration of the active substance along the various levels of the trophic chain)<sup>29</sup>.

## **Bioavailability**

Definition derived from ISO 17402. Degree to which chemical substances present in the environment can be absorbed or metabolised by an organism, or be available for interaction with biological systems. Bioavailability is specific to an organism and a contaminant, and depends on factors related to exposure time, transfer of contaminants from the medium to the organism, their accumulation in the organism and their subsequent effects. Bioavailability is therefore approached here as a dynamic process, which can be described by the following three successive phases:

- the availability of the contaminant in the environment, called 'environmental availability', which corresponds to the fraction of the compound potentially available to organisms in the environment;
- the absorption of the contaminant by the organism, also called 'environmental bioavailability', which corresponds to the fraction of the compound effectively available in the environment that an organism has taken up (uptake) by physiological processes;

29. Glossary of the Pesticides Properties Data Base (PPDB): <http://sitem.herts.ac.uk/aeru/ppdb/en/index.htm> (accessed 9/01/2023).

- the accumulation and/or effect of the contaminant on the organism, also known as ‘toxicological bioavailability’.

### **Biocontrol**

Defined according to the definition of the *Code rural et de la pêche maritime* (French Rural and Maritime Fishing Code) in CRPM, art. L.253-6: "Agents and products using natural mechanisms in the context of integrated pest management. They include in particular i) macroorganisms and ii) plant protection products comprising micro-organisms, semiochemicals such as pheromones and kairomones, and natural substances of plant, animal or mineral origin.

The list of biocontrol plant protection products covered by articles L.253-5 to L.253-7 of the Rural and Maritime Fishing Code is updated monthly and published online via a note from the Ministry of Agriculture and Food<sup>30</sup>. This note does not list macroorganisms but includes insect traps combining pheromones, food attractants or insecticides in a closed container.

### **Biodiversity (target/non-target)**

The widely accepted definition established in 1992 within the framework of the UN and the Convention on Biological Diversity (CBD) is used here: biological diversity is the "variability among living organisms from all sources, including, inter alia, terrestrial, marine and other aquatic ecosystems and the ecological complexes of which they are part; this includes diversity within species, between species and of ecosystems" (Article 2 of the Convention on Biological Diversity adopted at the Earth Summit in Rio de Janeiro in 1992).

This definition is also very close to the one used in the analytical glossary produced in the French Assessment of Ecosystems and Ecosystem Services (Efese)<sup>31</sup> in 2017, where biodiversity "refers to the variability of living organisms from all sources and includes diversity within species, between species and of ecosystems".

Beyond its taxonomic dimension, biodiversity is also considered from the perspectives of population dynamics, flows, interactions, ecological processes and ecosystem functions.

On the other hand, the concept of ‘non-target biodiversity’ proposed in the CSA referral has been omitted for the following two reasons. Firstly, the concept of target is associated with organisms and not with biodiversity. Secondly, it cannot be considered *a priori* or in a general manner. Indeed, it is relative to the intention of the user: what is not targeted by a given user in a given context is non-target. The distinction between target and non-target organisms can therefore only be made on a case-by-case basis.

30. <https://agriculture.gouv.fr/quest-ce-que-le-biocontrôle> (accessed 9/01/2023).

31. <https://www.ecologique-solidaire.gouv.fr/levaluation-francaise-des-ecosystemes-et-des-services-ecosystemiques> (accessed 9/01/2023).

### Co-formulant

A substance with no phytopharmaceutical activity of its own, which is incorporated into a plant protection product as a supplement to an active substance or adjuvant in order to facilitate the handling of the product, enhance the effectiveness of the active ingredient or increase the product's safety. These products are part of the commercial composition of a PPP. A list of co-formulants which are not accepted for inclusion in a PPP, adjuvant or combination product is available in Commission Regulation (EU) 2021/383 of 3 March 2021, amending Annex III to Regulation (EC) No 1107/2009 (2009b).

### Contamination

Understood in the manner used in the 2005 CSA 'Pesticides, Agriculture and the Environment' (Aubertot *et al.*, 2005a): the term contamination refers to "the abnormal presence of substances, micro-organisms, objects or living things. [...] (BRGM, Jeannot *et al.*, 2000). The definition of contamination involves the concept of the normality of the presence of substances in a given environment. However, this term does not include the manifestation of potential effects linked to this presence. [...] the presence of substances is considered polluting if it reaches a threshold for which damage is likely to occur".

### Ecological/ecosystem function

In the scientific literature, 'ecological function' (or ecosystem function, the preferred term in this CSA) is associated with definitions that can vary according to authors and their personal preferences. In this CSA, and largely guided by the definition adopted in 2018 by Brodie *et al.* (2018), an ecosystem function is defined as a set of activities and processes provided by a species or a group of possibly interacting species that contribute to the functioning of an ecosystem (e.g. maintenance of biogeochemical fluxes or pools, support for ecosystem productivity, regulation of interactions between two components of the ecosystem, prevention or limitation of the direct and indirect impacts of various environmental pressures).

Ecosystem functions are "at the heart of the relationship between the **biodiversity** of ecosystems and the production of **ecosystem services**" (definition proposed by CGDD in 2010 within '*Projet de caractérisation des fonctions écologiques des milieux en France*' (Project to Characterise the Ecological Functions of Environments in France). In this CSA, they are grouped into 12 categories (largely based on the 2010 CGDD report), associated with one or more **ecosystem services** that they support. By taking these ecosystem functions into account, the CSA aims to link our knowledge of the effects of **plant protection products** on **biodiversity** to all the activities or processes that are carried out by one or more types of organisms, and the consequences that this may have on **ecosystem services**.

## Ecosystem

The analytical glossary compiled as part of the Efese (French Assessment of Ecosystems and Ecosystem Services)<sup>32</sup> in 2017 led us to adopt the following definition of ecosystem: “A dynamic complex of plant, animal and microorganism populations, associated with their non-living environment and interacting as a functional unit.”

“The definition of the ecological status of ecosystems and its measurement must make it possible to express, in a readable way, the multiple dimensions of interest in the state of ecosystems and their biodiversity. It allows the risks of irreversible alteration of these ecosystems and their functioning to be documented and reflects their capacity to provide goods and services sustainably, to regulate their pressures, and to maintain their heritage aspects.”

## Ecosystem services

Conceptually, it was decided to use the framework proposed by the French Evaluation of Ecosystems and Ecosystem Services (Efese, 2017<sup>33</sup>). This defines ecosystem services as “socio-economic benefits derived by humans from the sustainable use of the **ecological functions** of ecosystems”.

This definition was further elaborated in the synthesis of the study on agricultural ecosystems<sup>34</sup> by clarifying the distinction between: **ecological function** (e.g. pollination of plants in general, whether cultivated or wild); ecosystem service, as a biophysical process impacting on human activity (e.g. pollination of crops); and ecosystem service benefit, as the value of the benefit derived from the service by one or more stakeholders, whether in monetary or non-monetary form. This distinction is important because different **ecological functions** may constitute the provision of an ecosystem service, and different stakeholders may derive different benefits from the same ecosystem service.

All services were considered, as listed in the latest version of CICES (Common International Classification for Ecosystem Services, version 5.1; Haines-Young and Potschin, 2018).

## Exposure

Contact between one or more pollutants and one or more organisms. A distinction is made between acute, subchronic and chronic exposure, depending on their duration and taking into account the life span of each species.

In this CSA, we have used the concept of exposure to mixtures in the case of simultaneous exposure to several plant protection products (including their possible **transformation**

32. <https://www.ecologique-solidaire.gouv.fr/levaluation-francaise-des-ecosystemes-et-des-services-ecosystemiques> (accessed 9/01/2023).

33. <https://www.ecologique-solidaire.gouv.fr/levaluation-francaise-des-ecosystemes-et-des-services-ecosystemiques> ; <https://www.ecologie.gouv.fr/sites/default/files/Thema%20-%20Efese%20-%20Le%20cadre%20conceptuel.pdf> (accessed 9/01/2023)

34. See *Sur la biodiversité* <https://www.inrae.fr/sites/default/files/pdf/efese-services-ecosystemiques-rendus-par-les-ecosystemes-agricoles-synthese-2.pdf> (accessed 9/01/2023)

**products** as well as **adjuvants** and **co-formulants**). This concept also includes exposure to multiple stressors. It may be used in the case of simultaneous exposure to one or more plant protection products and other types of chemical contaminants or other potential sources of stress, some of which may be related to climate change (e.g. increased average temperatures and their fluctuations, increased intensity of precipitation and drought, ocean acidification).

The concept of exposure allows the link between **contamination** and effect to be established.

### **Fate (of PPPs)**

The fate of substances in plant protection products and their **transformation products** is mainly determined by retention (adsorption, absorption, penetration, physical or chemical stabilisation) and degradation (biotic and abiotic) processes, which will determine their mobility and persistence, and therefore their environmental availability.

### **Focal species**

Species chosen to represent the most vulnerable species to the different pressures on a habitat, taking into account different parameters such as home range size, dispersal capacity, and degree of specificity of trophic resources needed.

### **Matrix**

The term is used in the sense of 'environmental matrix' to represent the various media in or on which **plant protection products** may be found. These matrices are distributed in different environments (terrestrial, continental or marine aquatic, atmosphere) and may be physical (soil, rainwater, surface or marine water, sediment) or biological (biota).

### **Plant protection products**

Understood in the sense established in the referral: "Products and organisms intentionally introduced for the stimulation of natural plant defences, the protection of crops and the management of non-agricultural areas (gardens, green spaces and infrastructures), including in particular herbicides, insecticides, fungicides, as sprays or seed coatings, basic substances, as well as biocontrol as defined in Article L.253-6 of the *Code rural et de la pêche maritime* (Rural and Maritime Fishing Code)".

Formulations (including **adjuvants** and **co-formulants**) and transformation products are taken into account.

All products present in the environment are included if they have been or are being used for crop protection purposes or for the maintenance of non-agricultural areas (including if they are misused). This includes those that are now banned in France, but which are still found (or their transformation products) in the environment because of their persistence.

### **Remediation**

Measures taken *a posteriori* to reduce the contamination of the environment (soil, water), by promoting degradation and retention processes, which thus reduces the **exposure** of organisms.

### **Transfers**

Processes of exchange of **plant protection products** between different environmental or biological matrices. In the context of this CSA, this term includes transport by convection, diffusion and/or dispersion, including trophic transfer.

### **Transformation products**

After application, active substances can be degraded by biotic (e.g. biodegradation by microorganisms) or abiotic (e.g. photolysis, hydrolysis) processes into transformation products, depending on their characteristics and the physico-chemical conditions of the environment. Transformation products can accumulate and impact the organisms present in the various environments. They may, depending on the case, be more toxic, of equivalent toxicity, or less toxic than their parent molecule, with a mode of action that may be similar to or different from that of the latter.

### **Vulnerability**

Vulnerability, from organisms to ecosystems, can be described as the interaction between their level of exposure to anthropogenic pressures, their degree of sensitivity, and their capacity to recover, which in turn depends on the physiological state of individual organisms and their exposure to other sources of stress.

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# Working group

The names of the INRAE scientific departments are abbreviated as follows: *Sciences pour l'action, les transitions, les territoires* (Sciences for Action, Transitions and Territories): ACT; *Agroécosystèmes* (Agroecosystems): AgroEcoSystem; *Écosystèmes aquatiques, ressources en eau et risques* (Aquatic ecosystems, water resources and risks): AQUA; *Économie et sciences sociales* (Economics and social sciences): EcoSocio; *Santé des plantes et environnement* (Plant health and environment): SPE.

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**Sacha Desbourdes**, Graphic design.

## Monitoring Committee

Members: MTE/CGDD (**Gwenaëlle Hello**), MAA/DGER (**Bénédicte Herbinet** and later **Antoine Legal**), MESRI/DGRI (**Enrique Barriuso**), Ifremer DG (**Léa Marty** and later **Olivier Le Pivert**), INRAE DS Environnement (**Thierry Caquet**) and INRAE DEPE (**Guy Richard**).

Invited to attend meetings: MTES/DGPR, MTES/DEB, MAA/DGPE, MAA/DGAL, MSS/DGS, Anses, OFB, Écophyto/CSO Recherche et Innovation.

## Stakeholder Advisory Committee

**Participating organisations:** ACTA (Association de coordination technique agricole), AFA (Association française d'agronomie), Agences de l'eau, Axema (Union des industriels de l'agroéquipement), CNC (Comité national de la conchyliculture), CNPME (Comité national des pêches et élevages marins), Coop de France, Fédération nationale du négoce agricole,

FNAB (Fédération nationale d'agriculture biologique des régions de France), FRB (Fondation pour la recherche sur la biodiversité), Générations futures, IBMA France (Association française des entreprises de produits de biocontrôle), LPO (Ligue pour la protection des oiseaux), OPIE (Office pour les insectes et leur environnement), Plantes et Cité, Réseau Civam, Solagro, UFC-Que choisir, UICN (Comité français de l'Union internationale pour la conservation de la nature), UIPP (Union pour la protection des plantes) puis Phytéis, UPJ (Union des entreprises pour la protection des jardins et des espaces publics).

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As part of the Ecophyto II+ plan, INRAE and Ifremer were in 2020 commissioned to conduct a collective scientific assessment of the impacts of plant protection products on biodiversity and ecosystem services. The results, released in May 2022, confirm that plant protection products contaminate all types of terrestrial and aquatic environments, all of their compartments and most of their organisms.

Contamination varies according to the distance from the areas where the products are used, principally in agriculture. It causes direct and indirect impacts on ecosystems, notably the decline of populations of terrestrial and aquatic invertebrates and birds, and the loss of ecosystem functions and services. Different measures can help mitigate contamination and its impacts, with varying degrees of effectiveness depending on the particular combination of measures (regulations, conditions of use of products, etc.).

This assessment identifies research needs, in particular to improve our understanding of dynamic phenomena in a context of multiple environmental stresses. It aims to characterise and predict impacts and to improve the procedures for assessing the risks associated with the use of plant protection products.

This book is targeted at all stakeholders and decision-makers with an interest in the use of plant protection products, whether they are involved in the legal, political, industrial or non-profit sectors, or in research or teaching.

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