

European arthropods and their role in pollination: scientific report of their biodiversity, ecology and sensitivity to biocides. September 2022



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Version	Changes	

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# European arthropods and their role in pollination: scientific report of their biodiversity, ecology and sensitivity to biocides.

Reference: ECHA-22-R-05-EN ISBN: 978-92-9468-131-7 Cat. Number: ED-08-22-194-EN-N DOI: 10.2823/147199 Publ.date: September 2022 Language: EN

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## List of abbreviations

Standard term / Abbreviation	Explanation
A.S.	Active Substances
BB(s)	Bumble Bee(s)
ECHA	European Chemicals Agency
EFSA	European Food Safety Authority
EU COM	European Commission
FVI	Flower-Visiting Insects
HB(s)	Honey Bee(s)
NBP	Non-Bee Pollinators
PPBD	Pesticides Properties DataBase
PPP	Plant Production Products
SSD	Species Sensitivity Data

### Summary

Non-bee pollinators (NBP) are a group of species with very diverse ecology. Indeed, they inhabit various habitats, using these as nesting sites, for shelter, as source of food and as hunting ground for prey. Depending on the life stage of species, NBPs can inhabit different areas and their overall contribution to pollination can be diverse. For example, adult butterflies mainly forage on nectar, whereas their larvae are herbivorous. Based on the highly diverse lifestyle, NBP's exposure to biocidal products can take place over different environmental compartments or matrices and they can be more exposed during certain life stages in which they are more sensitive. Mainly, exposure can take place through consumption of contaminated food (e.g., nectar, pollen, leaves) and water (e.g., puddles, natural water bodies) and/or through contact to contaminated materials used for shelter or nesting sites (e.g., soil, mud, litter, wood, stems). Also, when products are sprayed and insects come in contact to them while flying, exposure in airspace can be assumed. As some biocidal products are used against fly larvae in manure or dung, exposure of species is possible, if they use dung or manure as food source (e.g. some adult butterflies) or as nesting sites. Hence, the NBP can get exposed to biocidal products according to different routes of exposure. This scientific report aims to collect the available information on NBP in order to facilitate future research on the effects of biocides on these organisms. To reach this aim, firstly, a literature review related to the ecology and the sensitivity to insecticides of Diptera, Lepidoptera, non-bee Hymenoptera (Symphyta), and Coleoptera was done. Furthermore, a collection of toxicity endpoints of NBP exposed to active substances has been conducted. Ultimately, a data set of 143 toxicity end points in arthropod pollinators has been gathered across nine active substances and further analysis has been conducted to establish whether their sensitivity significantly differs to that of the honey bees (HBs). Although the database is relatively scarce, it seems to indicate that some NBP species, at certain life stages (e.g., larvae), can be as sensitive or even more sensitive than HBs for some active substances. Nevertheless, the results shows that the development of a risk assessment approach on NBP would have to deal with a high heterogeneity and limited knowledge of sensitivity variability of these organisms. In conclusion, further research is needed to fill the current data gaps on the NBP ecological traits, which species and for which substances/mode of action they appear to be more sensitive to address the risk assessment of biocidal products.

## 1. Introduction

Nowadays, bees are well documented as effective pollinators of global crops of economic importance. However, the contributions by pollinators other than bees have been little explored despite their potential to contribute to crop production and stability in the face of climate change.

On 2 December 2019, the European Commission mandated ECHA to develop a guidance for assessing the risks to arthropod pollinators (including bees) from biocides exposure to ensure a high and harmonised level of protection of the environment, considering EFSA's Guidance Document on the Risk Assessment of Plant Protection Products on Bees (currently under review). In addition, ECHA was requested to specify the information required to enable a conclusion by the evaluating authority on whether products comply with the criteria under Article 19(1)(b)(iv) of the Biocidal Products Regulation concerning bees and other arthropod pollinators.

In order to fulfil the Terms of Reference from the mandate, a scientific expert group composed of experts from Member States with specific scientific competence in risk assessment to bees, other arthropod pollinators and bee biology with the support from experts from the European Food and Safety Authority (EFSA) has been set up by ECHA. Within the group, several experts have been focusing on the non-bee pollinators with the goal of ensuring that a risk assessment methodology will be available in the future to protect these organisms and therefore ensure that the ecosystem service of pollination they provide is sufficiently protected.

During the development of this report two consultations have been carried out to the ECHA's ad hoc stakeholder consultation group on pollinators guidance (experts from academia, industries, and NGOs) to ensure all relevant information with regards to non-bee pollinators (NBP) had been collected.

NBP include, among others, flies, beetles, moths, butterflies, wasps, ants, birds and bats. The first aim of this scientific report is to compile and analyse available studies on families of nonbee arthropod insects that are known to visit flowers and identify those ones have a role in pollination, focusing on their ecological traits, habitat types and feeding behaviour (chapter 3).

Once the key families of organisms were identified, the work focused on collecting available data by a scientific literature review on sensitivity of these organisms with the aim of understanding whether HB can be used as surrogate species to protect other NBP (chapter 4). Overall, a data set of 143 toxicity end points in arthropod pollinators has been gathered across nine active substances and further analysis have been conducted to establish, whether their sensitivity significantly differs to that of the HB, and thus would need to be considered in the environmental risk assessment in the future (chapter 4, section 4.2).

This report should form the basis to decide in the future whether a quantitative risk assessment for NBP is possible based on the currently available data and should serve as starting point for further guidance development. Furthermore, important data gaps are highlighted and recommendations for future research are proposed (chapter 4, section 4.3).

## 2. Definitions and scope

The above-mentioned mandate of the EU COM focuses on arthropod pollinators. In general, pollination is the act of transferring pollen grains from the male anther of a flower to the female stigma. However, most so-called pollinators have only been determined to be flower-visiting insects (FVI) because the usual visual observations on flowers are not sufficient to prove pollen deposition on the stigma (Uhl and Brühl 2019). FVI are defined as insect species that directly interact with flowers at least in the flying adult life stage (Wardhaugh 2015). Therefore, the term "flower-visiting insects" and the term "non-bee pollinators" will be used side-by-side. The focus of this scientific report is on non-bee arthropods, visiting flowers and their contribution to pollination.

This report does not aim at providing a risk assessment strategy nor to conclude on whether non-bee pollinators could be protected by the risk assessment strategies set up for HB. However, it aims to collecting the available information and state of the art with regards to these organisms to facilitate later discussions, research and guidance development in the future.

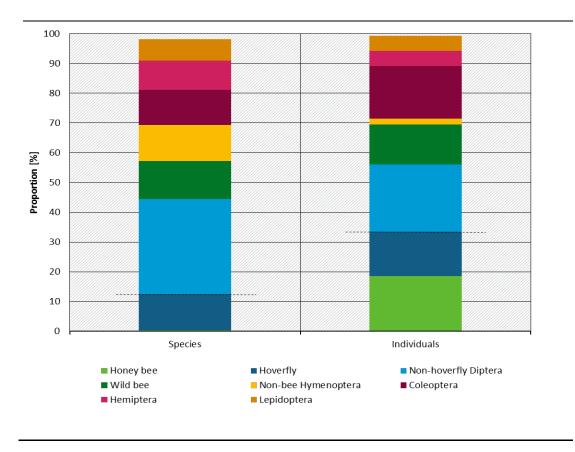
## **3. Ecological profile of flower-visiting insects and their** role as pollinators

FVI are an ecologically diverse group of mobile, flying species (Uhl and Brühl 2019) with representatives from the orders: Diptera (mainly dominated by flies), Lepidoptera (moths and butterflies), Hymenoptera (bees, wasps and ants), and Coleoptera (various beetle families) (Wardhaugh 2015). The group "bees", which includes *Apis mellifera* (the western honey bee), species belonging to the genus *Bombus* (bumble bees) and different families known as "solitary bees" (e.g., species belonging to the families Megachilidae and Halictidae), is addressed in the EFSA guidance (2012). Therefore, only non-bee FVI/pollinators are considered in this document.

Some of the above-mentioned families contain flower-visiting species, which are also pests, and therefore target organisms for biocidal products (e.g., ants, mosquitoes). Thus, it was decided to exclude ants and mosquitoes from the group of relevant FVI, supported by the fact that these families do not significantly contribute to pollination in Europe. Some further species/families are agricultural pests regarded as nuisances like housefly/stable fly, paper wasps and blow flies. However, Diptera and Hymenoptera are important flower visitors and pollinators. Therefore, it was decided not to disregard these species in this document. The same accounts for the herbivorous life stages of Lepidoptera.

A study from Wardhaugh (2015) estimates that more than a million arthropod species globally regularly visit flowers, mainly to find food, shelter or a mate. As a side effect, some of these species also contribute to pollination. The most abundant flower-visiting arthropods are from the "big four" insect orders, namely the orders Diptera (flies), Lepidoptera (moths and butterflies), Hymenoptera (bees, wasps, ants) and Coleoptera (beetles), (see e.g. Kevan and Baker 1983; Wardhaugh 2015; Uhl and Brühl 2019).

Recent research done by Grass et al. (2016) investigates flower visitations of insects in wildflower plantings situated in the central German agricultural landscape. Figure 1 summarizes the results and shows that in fact a diverse community of species are visiting flowers (UBA-Texte 54/2019). The study also shows, that aside from bees and hover flies, flowers were visited by a diverse community of other insect taxa. In fact, non-bee/non-hover fly insects made up more than half of the visiting individuals and accounted for more than 75% of FVI species (Figure 1).



Footnote: The dashed line shows the cumulative fraction of honey bee and hover fly flower visits

Figure 1: Contributions of different insect taxa to flower visitations in wildflower plantings in central Germany (UBA Texte 54/2019; adapted from Grass et al.,2016).

More research done by Rader et al. (2015) investigated flower-visits in crop systems and they found that the 38% of the flowers were visited by non-bee species. In addition, Wardhaugh (2015) presented an overview of species of the orders Diptera and Coleoptera, their reason on visiting flowers and if they are known or suspected pollinators (see supplementary data in Annex).

#### **Ecology**

Other than visiting flowers, in at least their adult stage, FVI differ substantially in their ecology (Ollerton 2017, cited in Uhl and Brühl 2019). While adult bees feed predominantly on nectar and their larvae mostly on pollen (Michener 2007), other FVI groups such as moths,

butterflies and beetles also have herbivorous life stages (Koch and Freude 1992; Ebert 1994; Scoble 1995, cited in Uhl and Brühl 2019). Similar to a large part of solitary bee species, there are also other FVI groups with soil-dwelling larval stages, for example flies and beetles (Koch and Freude 1992; Frouz 1999).

#### Habitat types

There are multiple habitat types that FVI species use throughout their life cycle according to specific functions at certain phases (Uhl and Brühl 2019) in their life cycle. Table 1 gives an overview of relevant habitat types used by NBP.

**Table 1:** Habitat types for NBP (Diptera, Lepidoptera, non-bee Hymenoptera, Coleoptera), adapted from Uhl and Brühl, 2019.

Habitat type	Life stages	Function
Airspace	Adults	Food search (foraging), mate search, nest search
Flowers	Larvae, adults	Food source (pollen, nectar), nesting, prey hunting
Other parts of plants (e.g. leaves, stems, twigs)	Larvae (herbivorous), pupae, adults	Food source, nesting, shelter
Soil	Larvae (soil- dwelling), adults	Nesting, prey hunting
Water (e.g. rivers, lakes, puddles), macrophytes	Larvae, adults	Food source, water consumption, nesting, shelter
Dung, manure	Larvae, adults	Food source, shelter, nesting
Other organic matter (e.g. litter, animal carcasses, faeces)	Adults	Food source

Depending on the ecological attributes of FVI species, relevant habitat types vary in time and space. In general, airspace, different plant parts (e.g. flowers, stems, leaves), soil and different natural (e.g. rivers, lakes) and artificial (e.g. puddles) water sources are recognized as relevant habitat types for FVI by Uhl and Brühl (2019).

#### Exposure pathways

Considering the diverse habitat types used by FVI during their life stage also their exposure pathways to biocidal products vary according to different compartments used during their life cycle. Table 2 gives an overview of the NBP relevant exposure pathways.

**Table 2:** Exposure routes and compartments/matrices of concern for life stages of FVI.

FVI order	Life stage	Exposure route	Compartments/ma trices of concern
Two-winged flies (Diptera)	Larvae	Oral	Plant structures (e.g. leaves, stems, twigs); dung, manure
		Contact <sup>1</sup>	Plant structures (e.g. leaves, stems, twigs); dung; manure; air; water; soil
	Adults	Oral	Nectar (floral and extra-floral), pollen; soil; water, macrophytes; dung, manure; other organic matter (e.g. litter, other insects)
		Contact	Plant structures (e.g. leaves, stems, twigs); nectar (floral and extra- floral); pollen; dung, manure; air; soil; water, macrophytes
Moths and butterflies (Lepidoptera)	Larvae	Oral	Plant structures (e.g. leaves, stems, twigs)
		Contact <sup>1</sup>	Plant structures (e.g. leaves, stems, twigs); soil; air
	Adult	Oral	Nectar (floral and extra-floral), pollen; puddles formed from dung, manure;-other organic matter (e.g. animal carcasses)
		Contact	Nectar (floral and extra-floral), pollen; plant structures (e.g. leaves, stems, twigs); air; dung, manure
Non-bee Hymenoptera (wasps, sawflies)	Larvae	Oral	Plant structures (e.g. leaves, stems, twigs)
		Contact <sup>2,1</sup>	Soil; man-made habitats (e.g. wooden buildings <sup>2</sup> ); air
	Adults	Oral	Nectar (floral and extra-floral), pollen
		Contact	Plant structures (e.g. leaves, stems, twigs); air; soil; man-made habitats (e.g. wooden buildings <sup>2</sup> )

Beetles (Coleoptera)	Larvae	Oral	Plant structures (e.g. leaves, stems, twigs)
		Contact <sup>1</sup>	Air; plant structures (e.g. leaves, stems, twigs); water; soil
	Adults	Oral	Nectar (floral and extra-floral), pollen; plant structures (e.g. leaves, stems, twigs); water
1		Contact	Nectar (floral and extra-floral), pollen, plant structures (e.g. leaves, stems, twigs); water; air; soil

<sup>1</sup> also relevant for pupae

<sup>2</sup> relevant for PT 08 products (Wood preservatives)

It can be deducted that FVI exposure to biocides through airspace, pollen and nectar, stems/leaves, soil, and water sources (rivers/lakes, puddles, guttation water) can subsequently lead FVI to exposure by direct overspray or spray/dust drift, by consuming contaminated food items such as pollen and nectar, stem or leaf material (e.g. lepidopterans, beetles) and contaminated water sources. Furthermore, they can be exposed by collecting nesting materials or by digging their nests in soil (e.g. beetles, flies) (UBA-Texte 54/2019). Not only herbaceous plants are attractive for bees and other pollinators, but also woody plants. Shrubs and trees can produce large numbers of flowers and are therefore contributing to the availability of nectar/pollen for arthropods (Mach and Potter 2018). This is supported by the results of the study done by Mach and Potter (2018), where they came up with a bee attractiveness rating for flowering plants, shrubs and trees. As a conclusion, they identified many species of flowering trees and shrubs that are highly attractive to bees. Supposedly, these plants could also be attractive to other pollinators. Also, non-attractive crop fields might be habitats for FVI due to weedy undergrowth consisting of flowering wild plant species. As reviewed by Uhl and Brühl (2019), linking habitat and plant protection products (PPP) contamination therein to individual exposure of FVI is difficult. Whereas there is some knowledge on food uptake and water intake by bees, data on the food spectrum of other FVI is scarce. Food intake is highly variable between species, hence single-species estimates cannot be generalized (Müller et al. 2006).

Overall, flower visiting insects encompass many ecologically important species that differently contributes to pollination. Indeed, FVI are a highly heterogeneous group as they use various habitat, and they contribute to pollination differently. Due to this heterogeneity, also their exposure routes to biocides and compartments of concern differ among FVI. In the recent years, knowledge regarding the benefits provided by insect pollinators is growing. However, there is limited information regarding how FVI, which represent a large proportion of insect, can benefit the entire biodiversity, and provide pollination services along with additional ecosystem services, such as the control of crop pests. The following chapter describes in more detail the biodiversity and the ecology of the four order Diptera, Lepidoptera, Coleoptera and Hymenoptera, that together cover the majority of FVI, focusing on their role as pollinators.

## 3.1 Order Diptera

#### 3.1.1. Main characteristics of relevant species

This diverse taxonomic group of globally over 150000 species is recognised as the second most important flower visitors (Larson et al. 2001; Winfree et al. 2011). FVI species are found in three families: Bombyliidae (bee flies), Syrphidae (hover flies/syrphid flies), and Tachinidae (tachinid flies). Hover flies are considered the key group (ca. 800 species in Europe) where nearly all species' adults consume nectar and sometimes also pollen (Larson et al. 2001; Winfree et al. 2011). However, this statement might need re-evaluation since recent findings by Grass et al. (2016) and Orford et al. (2015) show that a substantial part of FVI species in wildflower plantings and farmland are dipterans other than hover flies (UBA-Texte 54/2019).

#### Bombyliidae (bee flies, humbleflies)

Bombyliidae are one of the largest families of Diptera. This family comprises about 6,000 species, widely distributed in the northern hemisphere (Kastinger and Weber 2001). They are found on all continents except Antarctica and many oceanic islands, but Bombyliid species are typically and most frequently encountered in arid areas (Evenhuis and Greathead 2015). In Europe, this family is represented by more than 330 species (Pape et al. 2015). Bombyliidae are diverse in terms of size, ranging from barely from 1 mm to 20 mm in length. Adults are usually brown or grey. Their body is typically furry, which is very characteristic of this group, or covered with scales (Figure 2). Most genera are composed of broad-bellied species, although some have a narrow, conical abdomen (Aguado Martín et al. 2017). When flying, they make a sound, which, together with their hairy aspect, makes this group resemble to bees or bumble bees. As members of the order Diptera, they possess a unique pair of wings, large eyes, long and thin legs and very short antennae. This group is characterized by their flying skills, staying in the air without moving or flying very fast and agile.



**Figure 2:** Bombylius major (L., 1758) Large bee fly. source: Christian Kantner

Larvae are predators or parasitoids of other insects. Adults typically feed on nectar and pollen, behaving as important pollinators. They play a major role to ecosystems contributing to cross-pollination of plants and, in some cases, conservation of endangered species depends on bee fly pollination (Evenhuis and Greathead 2015). Very often, they present a long tubular feeding and sucking structure (proboscis) to sip flower nectar. Unlike butterflies, bee flies hold their proboscis straight, and cannot retract it.

### Syrphidae (hoverflies)

Syrphidae are a cosmopolitan family distributed worldwide, except in deserts and high latitude regions. Adults are among the most abundant and conspicuous Diptera (Vockeroth and Thompson 1987), and, in Europe, this family is represented by more than 800 species (Pape et al. 2015).

Hoverflies, also called flower flies or syrphid flies, are diverse in terms of size depending on the species. In general, they range from small (4 mm) to relatively large individuals (25 mm). They are usually black, very often with yellow or orange markings on head and thorax and particularly on the abdomen (Figure 3), more rarely predominantly brown, yellow, metallic green, or blue, or with various combinations of these or other colours (Vockeroth and Thompson 1987). Hoverflies of the family Syrphidae often mimic Hymenoptera.



Figure 3: Episyrphus balteatus (De Geer, 1776) Marmalade hoverfly. source: Nancy Ludwig

Larvae have a wide variety of habitats and food source, feeding on fungi, plant tissue or even other insects (Vockeroth and Thompson 1987). Adults are significant pollinators of many plant species, as most members of this family are FVI and feed on pollen and nectar (Innouye et al. 2015).

#### Muscidae (houseflies/stable flies)

Worldwide, the family Muscidae include approximately 9,000 species within 190 genera (Moon 2002). Muscidae are a family within the suborder Brachycera and, in Europe, there are about 45 genera and 575 species (Oosterbroek 2006).

Muscidae are small to large in their size (2-18 mm), usually coloured grey and black, some species show yellowish brown or extensive green to blue metallic coloration (Oosterbroek 2006). Muscidae are a richly shaped family, some typical blowfly-like (Chinery 2012). The mouthparts are well developed (Oosterbroek 2006), the antennae are 3 segmented and the second antennal segment is distinctly grooved (Watson and Dallwitz 2003). Ocelli are present, the eyes are mostly asymmetric, sometimes connected above the antennae or nearly closed (Watson and Dallwitz 2003). The thorax shows dorsal suture continuous across the middle with well-defined posterior calli (Watson and Dallwitz 2003). The scutellum is bare underneath and the subscutellum is absent (Oosterbroek 2006). The abdomen consists of 3-5 segments and are visible (Watson and Dallwitz 2003).

The larvae are carnivorous (Phaoniinae, Mydaeinae, Coenosia, Limnophora, Lispe, Graphomya), saprophagous in decomposing organic material (excrement, vegetable or animal), the third larvae instar may be also carnivorous (Muscini, Stomoxyni, Hydrotea) and rarely phytophagous (Atherigona) (Oosterbroek 2006). Many species are related to the human environment, the most commonly known species is *Musca domestica* (d'Assis-Fonseca 1968; Watson and Dallwitz 2003). The adult stages of most species are visiting flower and feed on nectar, some species are predators (Coenosiinae), feeding on blood (Stomoxyni) and wound liquor (Hydrotaea) and are therefore vectors of diseases (Oosterbroek 2006). Some species also eat pollen and use flowers as sites for predatory activity (Inouye et al. 2015). Some species are predators on other fly larvae like flower flies, crane flies and midges (Skevington and Dang 2011). Floral visits can be quite long up to 15 min for the pollen eating genus Thricops (Elvers 1980). The survey by Clement et al. (2007) showed that Muscidae are efficient pollinators for the plant genus *Allium*. Muscidae are also known as pollinators for brood-site deception including the mimicking of dung and carrion (rev. by Jürgens et al. 2013; Urru et al. 2011) but also important for the pollination of non-mimetic flowers (Orford et al. 2015).

#### Nemestrinidae (tangle-veined flies)

Most of the species of the family Nemestrinidae are found in the tropics and subtropics, especially in South America and Australia (Narchuk 2006). The family covers around 250 species worldwide in 23 genera (Woodley 2009). In Europe, 6 genera with 13 species are spread (Oosterbroek 2006).

Nemestrinidae are large flies, with a size range of 10 to 18 mm, frequently have a long proboscis with narrow sucking lobes and a densely pubescent body (Narchuk 2006). The head can be wider than the thorax with very large eyes (Bernardi 1973; Woodley 2009). The body is variable coloured, sometimes the thorax and the abdomen are banded, the wings are elongated and with complete venation (Oosterbroek 2006).

The larvae are endoparasitoids of beetles (family Scarabeidae) and grasshoppers. The host is still alive until the fourth larvae stage develops (Narchuk 2006; Oosterbroek 2006). Pupation takes place outside of the host's body (Narchuk 2006). The adults hover in the air over flowers and frequently visit flowers (Oosterbroek 2006). Since the adults are frequently covered with pollen, they are considered important for cross-pollination of plants (Narchuk 2006), especially

for plants with long tubes.

#### Stratiomyidae (soldier flies)

Worldwide, around 380 genera with 2,700 species are described (Zhang 2011). In Europe, 27 genera can be found with 140 species (Oosterbroek 2006).

Many of the species are brightly coloured with a size range of 3 to 20 mm (Woodley 2001). The shape is mostly slender to stout, sometimes flattened (Oosterbroek 2006). The antennae are divided into 7-10 segments, sometimes with swollen basal segments, the mouthparts are very often short, the wings are often clear with a distinct venation (Oosterbroek 2006), however the ability to fly is often not well developed (Chinery 2012). The feet have 3 lobes at the end (Chinery 2012).

The larvae are not predators or parasitoids, which is in contrast to many other families of the Diptera (Oosterbroek 2006). The aquatic larvae feed on algae and rotting material and the habitats are stagnant (genera *Oplodontha*, *Odontomyia*, *Stratiomys* and *Nemotelus*, see Woodley 2001) or running waters (genus *Oxycera*, see Woodley 2001), moist rock faces up to marshes and saline environments (Oosterbroek 2006). Terrestrial larvae can be found under different rotting material like leaves in the topsoil (genera *Beris*, *Sargus*, *Chloromyia* and *Microchrysa*, see Woodley 2001). Some species also dwell in ant nests (Woodley, 2001). Adults feed on honeydew and nectar and frequently visit flowers, however some species also hunt insects under the bark like dark beetles (Skevington and Dang 2011). Typcial habitats cover woodland, dunes, coastal habitats and always in the vicinity of water (Oosterbroek 2006).

#### Tabanidae (horseflies)

Worldwide, there are around 4,400 species distributed (Pape 2011). In Europe, 13 genera with 220 species are spread (Oosterbroek 2006).

Tabanidae have a massive physique and can fly very fast (Chinery 2012). The eyes are very large and often have brightly coloured patterns (Chinery 2012). The size is about 6 to 30 mm, the body is generally brown coloured with patterns and sometimes metallic (Oosterbroek 2006). The antennae have 6 or more segments, usually the third segment is very large, and the legs are powerful (Oosterbroek 2006).

The larvae are aquatic, semi-aquatic or terrestrial, mostly predators (on worms, snails and other larvae of dipterans) but also facultative saprophages (Oosterbroek 2006). The female adults are bloodsuckers to cattle and humans and can also transmit diseases, the males suck nectar (Chinery 2012). Many species of the family Tabanidae live in wetland soils and in beds of fast flowing streams (Skevington and Dang 2011).

#### Conopidae (thick-headed flies)

Worldwide, about 800 species in 47 genera are described. In Europe, 14 genera with 85 species are distributed (Oosterbroek 2006).

Conopidae are small (3 mm) to large (20 mm) with a slender to strong body shape (Oosterbroek 2006). Often the species are yellow-black patterned, and the mouthparts are conspicuously long (Oosterbroek 2006) and double kneed (Arnold and Jentzsch 2016).

The larvae develop parasitically in bumble bees (BBs), bees and wasps. The injection of the eggs is carried out in flight with special structures on the abdomen (Oosterbroek 2006). Some species

are parasitoids of honey bees (HBs), ant flies and cockroaches (Skevington and Dang 2011). The adults often visit flowers and feed on nectar, species with long mouths, for example, visit long-cup flowers (Oosterbroek 2006). Many different flowering plants are often visited, as a study by Flügel (1999) suggests. They are therefore considered important pollinators for different wild plants. The adults usually prefer warm conditions and can be found in flower-rich vegetation, such as near blooming heather or on trees and shrubs in flower (Oosterbroek 2006).

#### Calliphoridae (blow flies)

In Europe, 22 genera with 115 species are spread. The body is robust and their size ranges from 4 to 16 mm (Oosterbroek 2006). Many species are green or blue metallic colored with a silvery or golden shine. Head, body and legs are often heavily bristled (Figure 4).

The adults and larvae of Calliphoridae are the most common species found in carrion communities (Baz et al. 2007). Larvae of most Calliphoridae may also infest the bodies of living human or vertebrate animals and feed on the host's dead or living tissues (Oosterbroek 2006). Some species are predators or parasitoids of developing stages of Hymenoptera, Orthoptera and Isoptera (Skevington and Dang 2011). Adults often can be found on flowers, detritus, carrion and dung.



Figure 4: Musca caesar (Linnaeus, 1758; Macquart, 183) Blow fly. source: Christian Kantner

#### Tachinidae (caterpillar flies)

Worldwide around 8,500 species are known (O'Hara J.E 2013). In Europe, 880 species are described (Oosterbroek 2006).

The species have a body length of 2-20 mm (Oosterbroek 2006.) Most of the species are grey to black in their coloration and some species have yellow and red spots on the abdomen (Figure 5). Some species are metallic green or yellowish grey. Many species have a high number of bristles on their body (Oosterbroek 2006).

The larvae stages are endo-parasitoids of different insects and arthropods (Stireman et al. 2006). Some species deposit the eggs directly on the host and others on leaves and get eaten by caterpillars (Oosterbroek 2006). Adults can be found on walls, leaves and other parts of plants, however only few species are known to visit flowers (Oosterbroek 2006) such as adults from the tribe Phasiinae and Tachinini (Stireman et al. 2006). The importance for pollen and nectar as diet is poorly known (Stireman et al. 2006).



Figure 5: Eriothrix rufomaculata (DeGeer,1776) Tachinid fly. source: Nancy Ludwig

## 3.1.2 Habitat Types

The order Diptera has successfully colonized all continents and all habitat types except the open sea and the inside of glaciers (Courtney et al. 2017). However, species of midges (family Chironimidae) are breeding near the coast of Antarctica and are therefore the most southern living insects (Skevington and Dang 2011). The larvae can be found in various terrestrial and aquatic habitats (Teskey 1976; Ferrar 1987; Hövemeyer 2000; Courtney and Merritt 2009). Many larvae need a moist to wet environment - from living in organisms or tissue in plants and organic material up to contact with bodies of water (Courtney et al. 2017). Some larvae of stiletto flies (family Vermileonidae) live in dry areas like beaches or deserts (Courtney et al. 2017). As shown in Table 3, adult flies can be found on various habitats from dry to wetland sites, from rural to urban areas. Many fly species, especially within the family Muscidae, are associated with humans.

Table 3: Typical	habitat types of Diptera	a in different life stages	and their function.
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Habitat type	Life stages	Function		
Flowers	Adults	Food source (pollen, nectar), nesting, prey hunting		
Other plant structures (e.g. leaves, stems, twigs)	Larvae (herbivorous), pupae, adults	Food source, nesting, shelter		
Soil	Larvae (soil-dwelling), adults	Nesting, prey hunting		
Water, macrophytes	Larvae, adults	Food source, water consumption, nesting, shelter		
Dung, manure	Larvae	Food source, nesting		
Other organic matter (e.g. animal carcasses, fungi, other insects)	Larvae, adults	Food source, nesting		

### 3.1.3 Ecological role

According to Skevington and Dang (2011) Diptera have a high impact on ecosystems because of their diverse feeding habits. Most of the terrestrial larvae are decomposers of organic material and are important for the function of soils and providing nutrients for plants. Other species of Diptera (larvae and adults) are predators or parasitoids and parasites. Diptera are also important vectors for diseases and contribute to the propagation of plants and many pathogens like nematodes, bacteria and viruses. When it comes to transmission of diseases, midges are the most prominent family.

#### 3.1.4 Feeding behaviour

Flowers with nectar and pollen are important food sources for many fly species (table 4) and relevant for their flying activities (Skevington and Dang 2011). Flowers are also important for finding mates, mating and are searching sites for oviposition (Larson et al. 2001). Many fly families are recorded to visit flowers (Larson et al. 2001). The most well-known flower feeders and important pollinators are Syrphidae (Ssymank et al. 2011), but other families might be important too. However, the documentation of a clear pollination relationship is not clear in the scientific community (Larson et al. 2001). There is some knowledge about numerous families of Diptera, which visit flowers, examples are Bibionidae, Mycetophilidae and Culicidae (among Nematocera), Syrphidae, Bombyliidae, Conopidae, Stratiomyidae, and Nemestrinidae (among lower Brachycera); and among the higher Brachycera (Cyclorrhapha), the Muscidae, Anthomyiidae, Tachinidae and Calliphoridae (Kastinger and Weber 2001; Larson et al. 2001; Rotheray and Gilbert 2011).

	Bomb.	Syrphidae	Muscidae	Neme.	Stra.	Taba.	Cono.	Call.
A	Yes	Yes	Yes <sup>1</sup>	-	Yes <sup>2</sup>	Yes <sup>3</sup>	Yes	No
L	No	No	No	No	No	No	No	No

Table 4: Feeding behaviour of Diptera families. Does the family only feed on nectar or pollen?

(A=adults, L=larvae, Bomb=Bombyliidae, Neme=Nemestrinidae, Stra=Stratiomyidae, Taba=Tabanidae, Cono= ConopidaeCall=Calliphoridae)

The challenge is to understand the spectrum of the life cycle and ecological niches of most dipteran species, the knowledge thereby is scarce (Raguso, 2020). The analysis of visitation networks and pollen transport by Orford et al. (2015) strongly proposes that it is inappropriate to exclude non-syrphid Diptera from pollination studies. The authors suggest focussing on dipterans that might fill the niche of declining bees (Orford et al. 2015).

Although it is not clear which families of flies are important for pollination, the data show that flies visit various species of flowering plants and will be considered in the risk assessment for non-arthropods for biocides.

## **3.2 Order Lepidoptera**

### **3.2.1 Main characteristics of relevant species**

The Lepidoptera (moths and butterflies) is a species-rich order of the insects. Estimates suggest there are ca. 160,000 described species, whereas the total number of extinct species will be about half a million (Kristensen et al. 2007). The following paragraphs highlight some families, known as pollinators.

#### Sphingidae (hawk moths)

The family of hawk moths is most diverse in tropical regions, but they can be found throughout the world with about 1,000 species, of which 63 are present in the Palaearctic Region (Akkuzu et al. 2007).

Hawk moths are considered to be the main pollinators for flowers characterised by long, narrow, tubular corollas or spurs that restrict access to the nectar foragers. Hawk moths have two principal biological adaptations that enable them to be efficient pollinators of such flowers. They have tongues (probosces) that are long and thin, allowing them to drink nectar from long, narrow flowers. They are also one of the specialist nectar-feeders to have developed a hovering flight, similarly to glossophagine bats and hummingbirds (Kitching 2002).

<sup>&</sup>lt;sup>1</sup> most species, some are predators and blood-feeders

<sup>&</sup>lt;sup>2</sup> some species also hunt insects

<sup>&</sup>lt;sup>3</sup> males suck nectar, females blood

#### Zygaenidae (burnet moths)

Burnet moths are a diurnal moth family with a similar biology to many butterflies. They are often restricted to small areas as habitats. Adults are frequent flower-visitors, where they feed on nectar. On some occasions, burnet moths have been noticed to be the dominating pollinators of rare orchids. The distribution of many European species is not fully known as of now, and other knowledge on them is also limited (Cengiz et al. 2018).

Burnet moths mainly use dry grasslands as their habitats, though one of the most important habitat types for them is semi-natural pasture (Franzen and Ranius 2004). Moths of the family Zygaenidae are excellent indicators of environmental conditions (Cengiz et al. 2018).

#### Hesperiidae (skippers)

Skippers are commonly known by their quick, darting flight habits. Most skippers are brown or grey with a notable uniformity (Wang et al. 2015). 47 species of skippers can be found in Europe. Adult skippers of most species feed on floral nectar, but some also take up nutrients from bird droppings. Larvae live in shelters made of leaf spun together or just folded over (Wiemers et al. 2018).

#### Papilionidae (swallowtail butterflies)

The family of swallowtail butterflies includes about 589 species (Heppner 2008), but only 15 species can be found in Europe. They are mainly tropical, and have larger wings compared to other insects, which makes them powerful fliers. The coloration and patterns on wings can be extremely rich and diverse (Collins and Morris 1985). Adults of all species visit flowers for nectar (Wiemers et al. 2018).

#### Nymphalidae (brush-footed butterflies, four-footed butterflies)

Distributed worldwide, butterflies of this family are especially rich in the tropics. Painted ladies (*Vanessa cardui*, Figure 6) are one of the most widely distributed species of butterfly and can be found on all continents except Antarctica and Australia. To date, 246 species have been discovered in Europe. They are highly variable, and there are more species in this family than in any other family (Wiemers et al. 2018).

Adult feeding behaviour depends on the species, where some groups primarily seek flower nectar, while others only feed on sap flows, rotting fruit, dung, or animal carcasses. Caterpillar appearance and behaviour vary widely (Wiemers et al. 2018).



Figure 6: Vanessa cardui (Linnaeus, 1758) The painted lady. source: Christian Kantner

#### Pieridae (whites and sulfurs)

Species of Pieridae have long been the subjects of ecological and evolutionary studies (Wahlberg et al. 2014). Adults of all species visit flowers for nectar, and there are 57 species of Pieridae butterflies in Europe (Wiemers et al. 2018).

These delicate species seem to be vulnerable to many factors, such as weather, predators, and host–plant defences. For instance, instars are frequently recorded as being attacked by parasitoids (Courtney 1986).

#### Lycaenidae (gossamer-winged butterflies)

Worldwide in distribution, this second largest family of butterflies (Mulé et al. 2017) has approximately 4,700 species, of which 130 can be found in Europe (Wiemers et al. 2018). Most adults visit flowers for nectar, but some harvesters feed on woolly aphid honeydew and some hairstreaks feed on aphid honeydew or bird droppings. Lycaenidae often have a mutualistic or parasitic relationship with ants (Mulé et al. 2017).

#### 3.2.2 Habitat types

Butterflies tend to be associated with open areas in the temperate zone (Winfree et al. 2011), and many lepidopteran species can be found in agricultural landscapes as they also inhabit hedgerows or fragmented areas between arable lands (Braak et al. 2018). The larvae (or caterpillars) of most lepidopteran species are herbivores and feed on plant material such as leaves roots, flowers, seeds, or fruits. While some species are rather restricted in their caterpillar food spectrum and rely on one or a few host plants, others can consume a wide variety of plant species. Since some Lepidopteran species also feed on crops during their caterpillar stage, they have been classified as agricultural pests (UBA-Texte 54/2019). Table 5 gives an overview of relevant habitat types used by butterflies.

Most of the lepidopteran species feed on non-crop plants. In their adult stage, numerous species feed on nectar and occasionally on pollen. The intake of nectar can improve longevity and reproduction and so, butterflies and moths are regularly observed flower visitors (Hahn and Brühl 2016). Since adult Lepidoptera visit a wide number of plant species as nectar sources, they might be of benefit to plant diversity due to pollination (UBA-Texte 54/2019).

Habitat type Life stages **Function** Flowers Adults Food source (pollen, nectar), nesting, prey hunting Larvae (herbivorous), pupae, Other plant structures (e.g. Food source, nesting, shelter leaves, stems, twigs) adults Soil Larvae (soil-dwelling), adults Nesting, prey hunting Larvae, adults Food source, water consumption, Water, macrophytes nesting, shelter Adults Puddles from dung, manure Food source Other organic matter (e.g. Adults Food source animal carcasses, faeces)

**Table 5:** Typical habitat types of Lepidoptera in different life stages and their function.

#### **3.2.3 Ecological role**

Moths and butterflies are a common and species-rich insect group. Approximately 180,000 lepidopteran species have been described worldwide and they account for approximately 10% of all known insect species (Hahn and Brühl 2016). Lepidoptera are one of the most studied arthropod groups, yet most of the research has focused on diurnal butterflies, which represent approximately 10% of lepidopteran species. The rest of the species are classified as moths and have often crepuscular and nocturnal lifestyles (UBA-Texte 54/2019). Most lepidopteran species are nectarivorous and only very few consume pollen (EFSA 2015). Butterflies are among the most noticeable animals due to their wing coloration, which is perhaps the most diverse in the animal kingdom (Giraldo and Stavenga 2008). Only limited information of the role of moth pollination is available, but their role as pollinators might be yet underestimated (Hahn and Brühl 2016).

The importance of Lepidoptera in the ecosystem is based on their role in nutrient cycling as they break down plant tissue in both larvae and adult stages, and they are prey for insectivorous predators, such as birds and parasitoids (Hoang et al. 2011). They are also valuable pollinators in ecosystems because butterflies and moths show diurnal, crepuscular, and nocturnal habits, and many of them visit flowers throughout an entire day. They also transport pollen across a range of distances and are a species-rich group of potential pollinators. Moths as flower visitors can pollinate a range of plant species, of which most are specialized for moth pollination (certain orchids, for instance). The role of moths as pollinators can at present still be underestimated because only a limited number of studies on moth pollination are available (Hahn and Brühl

2016). Lepidoptera, and especially their larvae, can also be pests to crops (Fernandes et al. 2013). Species such as *Pseudophilotes baton*, the baton blue, have been listed as a highly vulnerable species (UBA-Texte 54/2019).

### 3.2.4 Feeding behaviour

Flower-visiting species are mostly part of the moth families Noctuidae (owlet moths), Sphingidae (hawk moths), Hesperiidae (skippers) and the butterfly superfamily Papilionoidea (common butterflies). Generally, lepidopteran species are nectarivorous, only very few consume pollen (Table 6). Though some species are reported to also feed on bird droppings, sap flows, rotting fruit, dung, or even animal carcasses (Norris, 1936; Boggs, 1987). The larvae (or caterpillars) are herbivorous and feed on plant material such as leaves roots, flowers, seeds and fruits (Scoble 1995). As some lepidopteran species also feed on crops during their caterpillar stage, they have been classified as agricultural pests. However, Lepidoptera feed on non-crop plants (Scoble 1995) and their feeding behaviour is not independent in space and time from reproductive behaviour. Flowers are likely also important for finding mates, mating and oviposition (Altermatt et al., 2011). Several studies have shown that Lepidoptera act as pollinators, but overall, the knowledge on the role of butterflies and moths – and especially nocturnal moths – is limited (Hahn & Brühl 2016). Nonetheless, in temperate regions, these insects play a minor role as crop pollinators (Hahn & Brühl 2016).

**Table 6**: Feeding behaviour of Lepidoptera families. Does the family only feed on nectar or

	Sphingidae	Zygaenidae	Hesperiidae	Papilionidae	Nymphilidae	Pieridae	Lycaenidae
А	Yes	Yes	Yes <sup>6</sup>	Yes	Yes <sup>7</sup>	Yes	Yes <sup>8</sup>
L	No	No	No	No	No	No	No

(A=adults, L=larvae)

<sup>6</sup> most species, some also feed on bird droppings

<sup>7</sup> some feed on nectar, some on sap flowers, rotting fruit, dung or animal carcasses

 $^{8}$  most species, some harvesters feed on honeydew or bird droppings

## 3.3 Order Hymenoptera

The order Hymenoptera is comprised of the two suborders Symphyta and Apocrita, with 132 families belonging to 27 superfamilies and appr. 153,088 extant species (Aguir et al. 2013).

#### 3.3.1 Main characteristics of relevant species

#### Suborder Symphyta

The suborder Symphyta is paraphyletic and probably the most primitive group within the Hymenoptera (Aguiar et al. 2013; Malm and Nyman 2015). Symphyta include ca. 14 families with more than 8,300 species widely distributed around the world (Taeger et al. 2010). In Europe, this suborder is represented by 11 families which include ca. 1,400 species (Taeger et al. 2006). Symphyta are found in a varied number of habitats, including meadows and forests.

They are frequently found in mountainous areas with shrub and tree vegetation, in cool and shady areas such as riparian forests and hardwood and deciduous forests (Stefanescu et al. 2018).

Sawfly is a general term applied to most members of this group, as females typically have a saw-like ovipositor used to cut plant tissue for egg insertion. Symphyta are characterised by the absence of constriction (i.e "waist"), between the thorax and the abdomen (first and second abdominal segments). Typically, most adults are fly-like in appearance, as opposed to the wasp-like habitus of most other Hymenoptera (Smith 1993). Adults show feeding-related mouthpart specialization either for consuming pollen or for consuming floral nectar (Jervis and Vilhelmsen 2000).

Larvae of Symphyta resemble caterpillars (the larvae of Lepidoptera) and are responsible of considerable damage to plants (Smith 1993). However, they are easily differentiated because sawfly larvae have five pairs of prolegs located on abdominal segments 2–6, while true caterpillars have at most only four pairs of prolegs on abdominal segments. The prolegs of Symphyta do not have crochets, whereas those of Lepidoptera larvae do (Goulet and Hubert 1993).

These insects feed on a wide range of sources. Although a few species are parasitic, nearly all species have plant-feeding habits (Goulet and Hubert 1993). Larvae are phytophagous or xylophagous. Adults of many species take the nectar from flowers, many others eat pollen, while a few species feed on the petals and pistils of the flowers (Jervis and Vilhelmsen 2000, Wäckers et al. 2007). Some even bite the young stems and branches to suck the fluid. Relevant families of Symphyta are described below.

#### Cephidae (stem sawflies)

The family Cephidae contains about 100 species, most of which show an Eurasian distribution and are included in the Holarctic subfamily Cephinae (Budak et al. 2011; Goulet and Hubert 1993). In Europe, 9 genera with 42 species are spread (Taeger et al. 2006). It is a small family with a thin integument, usually black or dark coloured and commonly with narrow yellow bands on the abdomen.

The morphology of members of this family is regarded as intermediate between the hymenopteran suborders Symphyta and Apocrita: antennae with more than 16 segments, weak constriction between the first and second abdominal segments, the lack of cenchri and the rough area on fore wings (Aguado Martín et al. 2017; Budak et al. 2011).

They are known as stem sawflies (Hill 1987) as their larvae are internal plant feeders, specially inside grass stems or twigs of woody plants. Adults are commonly observed feeding on grass species but also on nectar from members of Cruciferaceae or Euphorbiaceae and more rarely on Umbelliferae and other plants.

### Megalodontesidae (serrate-horned sawflies)

This family is composed exclusively by the palearctic genus Megalodontes (Taeger et al. 2006) which includes about 40 species restricted to the temperate regions of Eurasia (Goulet and Hubert 1993). In Europe, there are 22 species and most occur in the Mediterranean region (Taeger, 2002; Taeger et al., 2006).

Adults of this genus are characterized by their big teeth or jaws, as well as by presenting most of their cuticle black, except for some yellow spots on the head and thorax, and abdominal rings of the same colour. Legs and antennae are in general orange or yellow with black lines (Aguado Martín et al., 2017).

Larvae feed on herbaceous plants. Adults are attracted mostly by yellow flowers like those from Compositae, Ranunculaceae, Cistaceae and Umbelliferae, and live in bogs or flooded meadows, in clearings near water courses as well as in other habitats characterized by a high degree of humidity (Aguado Martín et al., 2017).

#### Tenthredinidae (common sawflies)

This is by far the largest family of the Symphyta in general. There is controversy regarding the number of species worldwide. Some authors speak about 1,775 species (Schmidt et al. 2017; Taeger et al. 2010), while others raise that figure to more than 6,000 (Goulet and Hubert 1993). Most of these species are found in temperate regions of the Northern Hemisphere, being the dominant sawfly group in boreal and arctic regions (Goulet and Hubert 1993). In Europe, more than 1,070 species are cited.

They are commonly named as the 'common sawflies' and can generally be recognized by their long-divided antennae in 9-11 (7-12) segments, cylindrical body and well-defined scutellum (Aguado Martín et al. 2017; Quinlyn et al. 2019). Adults are usually black, but most often with strikingly bright coloured patterns of green, brown, yellow, red, or white (Goulet and Hubert 1993).

Larvae are phytophagous, often feeding gregariously on the leaves of trees, shrubs and herbaceous plants. They usually possess 6–8 pairs of abdominal prolegs which, unlike those of lepidopterous larvae, lack crotchets (Alford 2012).

Adults live most commonly only for a few weeks in spring and early summer, though some are found throughout the summer and fall (Goulet and Hubert 1993). In general, adults are FVI feeding on nectar and pollen. In case of female adults, during reproducing period, they also feed on other insects like flies, coleopterans and even other Symphyta individuals.

#### Argidae (argid sawflies)

Argidae is the second most species-rich family of Symphyta with ca. 920 known species worldwide and with most diversity occurring in tropical regions (Schmidt et al. 2017). This family has a widespread distribution in Europe and is represented by 68 species within 5 genera.

They are easily recognized by their characteristic antennae (three-segmented, the third segment very long and usually forked), and they often have preapical spurs on the middle and posterior pairs of legs. Adults usually have a black head and thorax (more rarely with an orange thorax) and orange abdomen, or with the integument of almost the entire body of a metallic blue or green colour, except for antennae and legs that can be orange or black (Aguado Martín et al. 2017).

The larvae are caterpillar-like and feed on foliage of many plants (Goulet and Hubert 1993). Adults of some species of this family not only feed on floral and extrafloral nectar, but also on pollen (Wäckers et al. 2007).

#### Cimbicidae (cimbicid sawflies)

The family Cimbicidae, which contains ca. 200 described species around the world (Vilhelmsen 2019) with holarctic distribution (except for the subfamily Pachylostictinae; Smith 1988). The family is present in Europe with 6 genera including 55 species.

They are generally robust and large, and adults of some genera resemble bumble bees. Adults are recognized by the distinctively clubbed antennae, the absence of a mesoscutellar appendage and abdominal terga usually separated by folds, causing the abdominal spiracles to be oriented ventrally (Vilhelmsen 2019). Little information is available on the diet of adults. Mainly, they are referred to feed on deciduous tree species such as Ulmus, Salix, Alnus and Betula (Quinlyn et al. 2019; Smith 1993) and floral tissues such as petals and stamens. Therefore, adults may therefore consume nectar as well as floral tissues (Jervis and Vilhelmsen 2000). In addition, adults (mainly females) have been recorded preying on insects, which often takes place on flowers (Jervis 2000).

Larvae are solitary and feed externally on plant tissue. At the end of the larval development, they form a cocoon on the host plant or in the ground in which they pupate (Vilhelmsen 2019).

### Suborder Apocrita

#### Crabronidae (digger wasps)

Crabronidae is the largest of the spheciform families with about 3,400 species around the world in two subfamilies (Brothers and Finnamore 1993). Their diet consists of nectar and pollen, but they also prey on other insects as food for the larvae. Their nests are often built in wood, plant stems or the ground (Ghaderipour et al. 2021). 816 species can be found in Europe (Schmid-Egger et al. 2018).

#### Sphecidae (thread-waisted wasps wasps)

Sphecidae are a cosmopolitan but mainly tropical family of 660 species in three subfamilies (Brothers and Finnamore 1993). Some species can also be found in Europe (Cetkovi et al. 2004). This family includes a broad range of behaviour, including parasitoid and primitive social behaviour. They often build their nests in mud or construct it in the ground, some use pre-existing cavities. Sphecidae prey on other larvae (Brothers and Finnamore 1993).

#### Chrysididae (cuckoo wasps)

Cuckoo wasps are a family of obligate brood parasites (i.e., parasitoids and kleptoparasites). The use of hosts varies among sawflies, wasps and bees, walking sticks and moths. The family includes about 3,000 species in four subfamilies (Pauli et al. 2018). Currently, about 490 species are known in Europe, but the number could be higher. Although all cuckoo wasps parasitize other insects, they differ in the way they use their hosts: the parasitoids feed directly on the host larvae or pupae, whereas the kleptoparasites mainly make use of the food items stored in the host brood cell. Both often result in the death of the host. Some species parasitize other wasps and solitary bees, whereas others attack sawflies. Some also visit flowers, but their role as pollinators is rather insignificant (Paukkunen 2018). Adults of many species of the family visit bee cells for nectar and pollen (Martynova and Fateryga 2015).

Most cuckoo wasps live in warm, sunny habitats with sun exposed dead wood and/or bare sandy patches that provide nesting sites for their hosts. They are often seen on walls of wooden buildings or on sandy patches searching for nests to invade. Log houses and other wooden constructions such as wood poles are also typical habitats (Finnish Environment Institute 2013).

In regions with cooler climates, cuckoo wasps usually occur patchily in forest edges, sandy shores, and various artificial habitats, such as gardens, dry meadows and road verges. Since the hosts of most cuckoo wasps are highly specialized predators or pollen feeders, cuckoo wasps as their natural enemies may be particularly vulnerable to environmental changes (Paukkunen 2018). Cuckoo wasps are still quite poorly known and there is much uncertainty on their taxonomy, distribution and biology. Several species of the family are considered threatened due to habitat loss (Finnish Environment Institute 2013).

Species of Vespoidea vary in behaviour. Their range includes parasitoids, parasitic, solitary, social and scavenger behaviour. Parasitoids tend to parasitize the larvae of Coleoptera or soil-nesting wasps and bees. Predators and scavengers prey on a variety of insects and spiders, with the female providing prey as food for the larva. Herbivorous species make use of seeds, pollen or fungi (Brothers and Finnamore 1993).

#### Pompilidae (spider wasps)

Spider wasps are a cosmopolitan but mainly tropical family comprising of 4,000 to 4,500 species worldwide. Adult wasps are often mostly black with red, white, or yellow areas or markings (Brothers and Finnamore 1993).

Spider wasps are solitary wasps, and their larvae develop in paralyzed spiders (Ghahari et al. 2014). Most of the species dig simple burrows in the soil to store the spider on which the egg is laid. They do not visit the nest after laying the egg but build other nests elsewhere (Evans and Shimzu 1996). They can be found on flowers where they forage for nectar (Pitts et al. 2006).

#### Tiphiidae (tiphiid wasps)

Tiphiid wasps are a cosmopolitan but predominantly tropical family including about 1,500 species in seven subfamilies (Brothers and Finnamore 1993). Three subfamilies are present in Europe, namely the Tiphiinae, Myzininae and Metochinae (Bogush 2007). Adults are mostly black, sometimes with yellow or red markings (Brothers and Finnamore 1993).

All species are solitary, and their biology and ecology are very similar to those of Scoliidae. The larvae are ectoparasitoids of the larvae of soil dwelling Coleoptera (Bogush 2007). Both males and females feed on nectar and honeydew (Oliver et al. 2006).

#### Scoliidae (scoliid wasps, mammoth wasps)

Scoliid wasps are a predominantly tropical family with about 560 species. In Europe, the family is represented by about 22 species (Olszewski et al. 2016). They are large, stout-bodied and often black with red, yellow or white patterns (Grissel 2007).

All species are solitary, and the larvae are ectoparasitoids of coleopteran larvae. The female wasps dig the ground for larvae to paralyze them and use them as a host for their egg (Olszewski et al. 2016). Adults feed on nectar (Barrat 2003) and they often visit flowers (Winfree et al. 2011).

#### Vespidae

There are 300 species of Vespidae in Europe, divided into four subfamilies: Masarinae (the pollen wasps), Eumeninae (the potter or mason wasps), Polistinae (the paper wasps) and Vespinae. Vespinae are social wasps that build aerial or underground nests made of carton, composed of several combs protected by an envelope (Rasplus et al. 2010). This subfamily includes the true hornets (the genius Vespa) and the yellowjackets (genera Dolichovespula and Vespula). Pollen

wasps and paper wasps are cosmopolitan but mostly tropical with around 4,000 species in six subfamilies.

Vespidae adults are usually brown or black, often with yellow or white markings (Figure 7). Most species are solitary, though some are (eu)social. In solitary species, the larvae are usually predatory on other insects, particularly caterpillars. Eggs are laid in a cell constructed and provisioned by the adult female (Brothers and Finnamore 1993). The larvae are rarely supplied with pollen and nectar. In social species, the larvae feed on masticated insects or on glandular secretions provided by the female wasps. Some species are kleptoparasites in the nests of social species (Agriculture Canada 1993). Social Vespidae forage for water, pulp, carbohydrates, and animal protein. When hunting, social wasps are opportunistic generalists (Richter 2000).

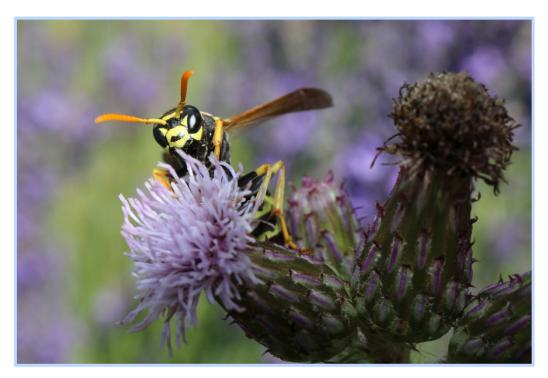


Figure 7: Polistes dominula (Christ, 1791) European paper wasp. source: Nancy Ludwig

*Vespinae* subfamily includes about 80 species in four genera. They are eusocial and construct annual or perennial compact paper nests, which are usually multicombed and covered with a paper envelope, suspended in the open, in sheltered positions or underground. The larvae are fed on masticated insects or even flesh from dead vertebrates (Brothers and Finnamore 1993).

*Masarinae and Pseudomasaris* subfamilies include about 250 species, all of which are solitary. Adult Masarinae are small to moderate in size with sessile metsoma. They nest in mud or soil, and provision the nests with pollen and nectar (Brothers and Finnamore 1993). Masarinae pollinate flowers as they visit them for pollen and nectar (Hunt and Toth 2017).

*Polistinae* subfamily includes about 700 species in about 25 genera. Most species are found in the neotropical region. They are eusocial and build annual or perennial paper nests, which are sometimes covered with a paper or mud envelope but are often exposed, suspended in the open or in sheltered positions. The larvae feed on masticated insects, especially caterpillars, but also on stored termites, ants or honey (Brothers and Finnamore 1993).

### 3.3.2 Habitat types

Parasitoid wasps (such as Sphecidae, Chrysididae, and Pompilidae) naturally need a habitat with a sufficient host population. Adult females of many parasitic wasps either aestivate or hibernate or do both. As shown in Table 7 they favour habitats such as grass tussocks, loose bark or dense and evergreen aerial vegetation (Shaw 2006). Man-made habitats, such as the walls of wooden buildings, also suffice when they search for host nests (Paukkunen 2018). Preferences for certain habitat sites vary between species (Shaw 2006).

Many wasp species nest in the ground. Nests can be made of paper, mud, sand, twigs and rocks. Some species, such as the Vespidae, construct compact paper nests, suspended in the open, in sheltered positions or underground. Parasitic species are also often seen on the walls of wooden buildings or on sandy patches searching for nests of their hosts (Paukkunen 2018).

Social wasps live in vegetated deserts, scrublands, different forests (dry, rainforest, high latitude/altitude coniferous forests) and arctic tundra (Hunt and Toth 2017).

Habitat type	Life stages	Function
Flowers	Adults	Food source (pollen, nectar), nesting, prey hunting
Other plant structures (e.g. leaves, stems, twigs)	Larvae, adults	Food source, nesting, shelter, prey hunting
Soil	Larvae, adults	Nesting, prey hunting
Man-made habitats (e.g. wooden buildings)	Larvae, adults	Host-nest foraging, nesting

**Table 7:** Typical habitat types of sawflies and wasps in different life stages and their function.

#### 3.3.3 Ecological role

The ecological role of Symphyta is linked to their high diversity and their behaviour as FVI. Many plant species serve as nectar and pollen sources to symphytan groups, but among the flowers that most attract adults are the Rosaceae, Umbelliferae, Ranunculaceae and Euphorbiaceae. Very often adults only visit the flowers of the host plant that feed their larval stages. Their great diversity also leads to a certain level of specialization and specificity in some species, determining even their morphological characteristics (Jervis and Vilhelmsen 2000). However, in most cases sawflies can only take nectar from wide flowers with fairly well exposed and easily accessible nectarines.

In relation with the larval stage of many symphytan groups, some sawflies and gall wasps are economically important pests of crop and ornamental plants. Larval stages feed on plant tissue and, very often, they feed on plant leaves. Certain species can be extremely damaging in certain types of crops (Smith 1993), but many of these have potential roles in the biological control of weeds as well. Notably, pest sawflies include both external foliage feeders such as the pine sawflies (Diprionidae), which can be major defoliators of coniferous forests, and concealed feeders such as the wheat stem sawfly, *Cephus cinctus* (Cephidae) (Quicke 2009).

Wasps are common flower visitors, especially the families Vespidae, Scoliidae and Pompilidae (Winfree et al. 2011). The family of Vespidae is also recognized as one of the main predators of phytophagous insects in natural environments and agricultural ecosystems (Bacci et al. 2009). Social wasps collect water, carbohydrates, fibres of plants and hunt arthropod prey or scavenge for animal proteins (Fernandes et al. 2013).

Wasps are also important regulators of arthropod populations, such as insect vectors of diseases and crop pests (Sumner et al. 2018). Moreover, wasps next to hoverflies, beetles, butterflies, bugs, and bees contribute to pollination, but the contribution of non-bee taxa are not well recognised (Rader et al. 2016). The social wasp *Vespula pennsylvanica*, the western yellowjacket, has even been observed to be a more effective pollinator than the honey bee (HB) in certain environments (Thomson 2019).

Social wasps occur in many different habitat types, and they hunt a wide range of prey. Their impact on the predation on other insects could be considered substantial (Hunt and Toth 2017). Parasitoid wasps also have an important role in natural and agricultural ecosystems, as they destroy eggs, larvae or cocoons of many other species of insects and arthropods. This can lead to other beneficial effects such as help to control invasive species (Penninsi 2010).

#### 3.3.4 Feeding behaviour

While larval stages of Symphyta are phytophagous or xylophagous feeding on plant tissue, the feeding habits of adult members of Symphyta include floral and extrafloral nectar as a sugar source. In the case of pollen feeding, it has barely been reported for some members of Symphyta (Table 8).

	Cephidae	Megalodontesidae	Tenthredinidae	Argidae	Cimbicidae
А	No	Yes	No	Yes	No
L	No	No	No	No	No

**Table 8:** Feeding behaviour of Symphyta families. Does the family only feed on nectar or pollen?

(A=adults, L=larvae)

Wasps have a diverse feeding behaviour from parasitic, solitary, social to scavenging behaviour. The diet of many adult wasps seems to consist of predominantly nectar or pollen. The larvae are mostly exhibiting carnivorous parasitic behaviour, and thus pollen and nectar have a minor part in their diet (Table 9).

Table 9: Feeding behaviour of wasp families. Does the family only feed on nectar or pollen?

	Crabronidae	Sphecidae	Chrysididae	Pompilidae	Tiphiidae	Scoliidae	Vespidae
А	Yes	No	Yes	Yes	Yes	Yes	Yes
L	No	No	No	No	No	No	No

(A=adults, L=larvae)

## 3.4 Order Coleoptera

The order Coleoptera is the most diverse and species rich insect group on earth with more than 380,000 species described (Zhang et al. 2018). There are 4 suborders (namely the Archostemata, Myxophaga, Adephaga and Polyphaga), 17 superfamilies and 168 families described (Lawrence and Newton 1982; Zhang et al. 2018 and supplementary information 1). Beetle species from multiple families are recognised as pollen feeders. However, data on the prominence of these taxa as FVI is scarce, and it has been estimated that approximately 30% of the global arthropod species are regular flower visitors. Therefore, several taxa might be quantitatively relevant FVI but cannot be recognised as such due to an insufficient database (EFSA 2015).

Beetles are mostly holometabolic, developing from eggs through several larval stages and pupation to the imago (Bouchard et al. 2017).

Among the nine largest beetle families, seven are (partly) phytophagous: the Curculionidae (true weevils), Chrysomelidae (leaf beetles), Cerambycidae (long-horned beetles), Buprestidae (jewel beetles), Scarabaeidae (scarab beetles), Tenebrionidae (darkling beetles) and Elateridae (click beetles) (Zhang et al. 2018). Recent research done by Weiner et al. (2016) characterized the abundance and number of flower-visiting and (possibly) pollinating insects within the framework of the Biodiversity Exploratories. Each exploratory contains 50 experimental grassland plots in Germany. As supplementary data (table S2), the number of species of collected Coleoptera are given as 49 for 119 different experimental grassland plots. The most common coleopteran families were the *Chrysomelidae* (with 18 different species), Elateridae (13), Curculionidae (13), Coccinellidae (9), Cerambycidae (10), Cantharidae (7), Apionidae (7), Oedemeridae (6) and Dastydidae (3). Two different species were recorded of the families Cleridae, Mordellidae, Phalacridae, Buprestidae, Rutelidae and Carabidae. One species each were found of the family of Bostrichidae, Cetoniidae, Malachiidae, Scaptiidae, Staphylinidae and Tenebrionidae.

Wardhaugh (2015) identifies coleopteran families which contain species that have been recorded visiting flowers and also indicates, for which of these families pollination is suspected or confirmed (see supplementary data, table S1). The following table summarizes these data, focusing only on the families known or suspected as pollinators. Furthermore, the table gives information on the reasons for visiting flowers. All in all, 25 families are pollinators, some of them (namely the Buprestidae, Mordellidae, Oedemeridae, Meloidae, Scraptiidae, Nemonychidae and Belidae) are obligate flower visitors (table 10, Wardhaugh 2015).

Table 10: Coleopterar	n families suspected or	confirmed as pollinators	(adapted from Wardhaugh 2015).
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Coleopteran family	Reason for visiting flowers
Hydrophilidae	Unknown
Ptiliidae	Unknown
Staphylinidae	Pollen/nectar/insect prey

Scarabaeidae	Pollen/nectar/flowers/deceived <sup>4</sup>		
Buprestidae	Pollen/nectar/flowers <sup>+</sup>		
Elateridae	Pollen		
Lycidae	Pollen/nectar		
Cantharidae	Pollen/nectar		
Dermestidae	Pollen		
Cleridae	Pollen/nectar/insect prey		
Melyridae	Pollen/insect prey		
Erotylidae	Pollen		
Monotomidae	Unknown		
Nitidulidae	Pollen		
Mycetophagidae	Unknown		
Mordellidae	Pollen/nectar <sup>+</sup>		
Oedemeridae	Pollen <sup>+</sup>		
Meloidae	Pollen/insect prey <sup>+</sup>		
Anthicidae	Unknown		
Scraptiidae	Unknown		
Cerambycidae	Pollen/nectar		
Chrysomelidae	Pollen/flowers		
Nemonychidae	Pollen <sup>+</sup>		
Belidae	Pollen <sup>+</sup>		
Curculionidae	Pollen/nectar/flowers		

The following paragraph gives an overview of flower-visiting beetle families. Due to a partly scarce data base, detailed descriptions could not be included for all families.

## 3.4.1 Main characteristics of relevant species

## Infraorder Scarabaeiformia

### Scarabaeidae (scarab beetles)

The Scarabaeidae (superfamily Scarabaeoidea, see Bouchard et al. 2011), commonly known as scarabs or scarab beetles, are a species-rich family including species which are ecologically and economically important as pollinators and agricultural pests (Moore et al. 2018). Worldwide, 1,900 genera and 27,000 species are described so far (Bouchard et al. 2017).

In central Europe, 42 genera with 211 species are described (Harde and Severa 2006). One important subfamily in Europe is the Cetoniinae (Figure 8) with well-known representatives being the flower beetles or rose chafers (e.g. *Cetonia aurata*, the green rose chafer). Adult Cetoniidae are known as nectar- and pollen-feeding flower visitors, whereas larvae develop in decaying vegetable matter or in plain soil (Krikken 1984; Ritcher 1958). Most species of Cetoniinae are diurnal (Ritcher 1958).



Figure 8: Oxythyrea funesta (Poda, 1761) The white spotted rose beetle. source: Christian Kantner

The family Glaphyridae (e.g. the genus Polypleurus, superfamily Scarabaeoidea) are known to forage and mate on mediterranean red, bowl-shaped flowers, e.g. poppies and red anemones (Keasar et al. 2010). Adults of glaphyrid beetles are often brightly coloured and hairy, often resembling bees or bumble bees (Keaser et al. 2010). Glaphyrid beetles are diurnal and active flyers, can often be seen hopping between flowers or foliage or flying (Keasar et al. 2010). The genus Pygopleurus shows a strong preference for red flowers, as could be shown by the experiments of Streinzer et al. (2019). In contrast, species of Trichopleurus (subgenus Eulasia) prefer violet, spiny flowerheads (e.g. *Onopordum* spp.) or yellow *Centaurea* spp. (both family Asteraceae) (Keasar et al. 2010).

Representatives of the Cyclophala, night active flower visitors, depend on volatile organic compounds to find their host plants (Maia et al. 2018). Flower-visiting species of the sub-family Rutelinae are e.g. *Phyllopertha horticola*, the garden chafer or garden foliage beetle, and *Hoplia argentea*, the gold dust leaf *beetle* (see results in Weiner 2016).

### Infraorder Cucujiformia

### Coccinellidae (ladybugs or ladybird beetles)

Native Coccinellidae (superfamily Cucujoidea, see Bouchard et al. 2011) amount to 104 species of 37 genera in central Europe (Harde and Severa 2006) and about 6,000 species placed in 360 genera worldwide (Tomaszewska and Szawaryn 2016; Bouchard et al. 2017). As is commonly known, adults and larvae are beneficial predators of aphids (Harde and Severa 2006), but some species complement their nutrition by feeding on pollen, guaranteeing survival and, at times, reproduction when prey is scarce (D'Ávila et al. 2016). Weiner (2016) reported 9 different species visiting flowers in grassland, the most prominent ones being *Coccinella septempunctata* (the seven-spot lady*bird beetle*), *Hippodamia variegata* (the Adonis ladybird beetle) and *Tytthaspis sedecimpunctata*, the sixteen-spot ladybird beetle. The seven-spot ladybird beetle is 6 to 7 mm in length and can be found in the palaearctic region (Bílý 1990).

#### Nitidulidae (sap beetles)

The Nitidulidae (superfamily Cucujoidea, see Bouchard et al. 2011) are a large family with more than 2,000 species (Ortloff et al. 2014). In central Europe, the family is represented by 154 species of 23 genera (Harde and Severa 2006). Sap beetles are mostly small, e.g. *Meligethes aeneus*, the rape beetle, is 1.5 to 2.7 mm in length, with a variety of body forms (Harde and Severa 2006). Lee et al. (2020) state, that the family Nitidulidae exhibit one of the most diverse feeding strategies of all beetles (including mycophagy, predation, saprophagy, necrophagy, autophagy, frugivory and tree sap/fluid feeding) and also occur in a variety of microhabitats (e.g. living and dead plants, leaf litter, subterranean fungi. Beetles of the genus Meligethes are feeding on pollen from different plant species (Romeis et al. 2005).

Rader et al. (2020) state two coleoptera families, the Coccinelidae and the Nitidulidae, as families visiting a wide range (> 12) of crops, also stating, that particularly Nitidulidae may also be of importance as agricultural pests.

#### Phalacridae (shining flower beetles, shining mold beetles)

The Phalacridae is a family within the superfamily Cucujoidea (Bouchard et al. 2011). There are 635 species and 52 genera described worldwide (Majka et al. 2008; Gimmel 2013), in central Europe 23 species from 3 genera are native (Harde and Severa 2006). The imagines are small, round-oval, shining and mostly black, with a domed upper side and can mostly be found on flowers (Harde and Severa 2006).

Species of the family occur nearly worldwide in terrestrial environments and are mostly feeding on fungi or are palynophagous (pollen-feeding) (Gimmel 2013).

#### Cerambycidae (long horned beetles)

The Cerambycidae is a family within the superfamily Chrysomelidae (Bouchard et al. 2011). They are one of the species-richest family of saproxylic beetles (est. 35,000 described species) (Peris-Felipo et al. 2011). According to Harde and Severa (2006), there are 247 described species and 90 described genera in central Europe.

A common representative in Europe is *Rutpela maculate,* the spotted longhorn. The imagines are 14 to 20 mm in length and can be found from May to August on flowers of mainly thistles and Apiaceae, foraging for pollen, others are known to feed on needles or twigs of healthy trees (e.g. *Monochamus* spp.; Rose 1957 cited in O'Neill et al. 2008). Larvae live in rotten or decaying wood (Bílý 1990).

### Chrysomelidae (leaf beetles)

The Chrysomelidae (superfamily Chrysomeloidea see Bouchard et al. 2011) are one of the most species-rich coleopteran families worldwide (more than 3,700 described species, belonging to more than 2,000 genera), with 2,300 species in the Euro-Mediterranean region (Magoga et al. 2018) and 595 species in 73 genera native in central Europe (Harde and Severa 2006). Leaf beetles are mostly phytophagous and partly known for their impact on agriculture (Magoga et al. 2018). They are facultative pollen eaters and important pollinators (Romeis et al. 2005). Most imagines are round or round-oval. Larvae can be found mostly feeding on leaves, roots or stems (Harde and Severa, 2006), adults are known to feed on leaves (Bieńkowski 2010).

A common representative in Europe is the leaf beetle *Cryptocephalus sericeus*. Imagines are 6 to 8 mm in length and shiny metallic coloured, mostly in green, violet or bronze and partly covered with hairs. Imagines prefer yellow flowers of Asteracea, e.g. dandelion and hawkweed (Bílý 1990).

### Cleridae (checkered beetles)

The Cleridae are a family in the superfamily Cleroidea (Bouchard et al. 2011). The checkered beetles (Cleridae and Thanerocleridae) contain apprx. 3,600 described species in 303 genera (Gerstmeier and Eberle 2011). In central Europe, 21 species and 11 genera are native (Harde and Severa 2006). The imagines have a metallic shine or are coloured, and the body is hairy (Harde and Severa 2006). The family Cleridae is a diverse group occupying different niches and relying on different food sources. Both adult beetles and larvae are predators and can be found on flowers or on wood, bones or on carrion (Harde and Severa 2006).

The subfamily Clerinae is the most specious subfamily of the family Cleridae (Gerstmeyer and Eberle 2011) and a flower-visiting species in Europe is *Trichodes apiaries*, the bee-eating or beehive beetle. Imagines can be found on flowers, whereas larvae feed on brood of solitary bees (Bílý 1990).

### Melyridae (soft-winged flower beetles)

The Melyridea (superfamily Cleroidea, see Bouchard et al. 2011) contain two important subfamilies, the Dasytinae and the Malachiidae. Some authors describe these subfamilies as families (see Harde and Severa 2006).

According to Harde and Severa (2006), there are 61 species and 14 genera of Malachiidae in central Europe. The imagines are small, elongate-oval beetles and 1.7 to 7 mm in length, rather brightly coloured and covered with moderately dense, erect and stiff hairs (El-Torkey et al. 2012). A common representative is *Malachius bipustulatus*, the malachite beetle. Imagines can be found from spring to late summer on flowers, where they prey on small insects, e.g. aphids or small diptera (Bílý 1990). Larvae inhabit various habitats (e.g. soil, leaf litter, under bark, in dead wood, in plant stems), where they are predators of xylophagous insects (El-Torkey et al. 2012).

The second subfamily, the Dasytinae, are described with 34 species belonging to 8 genera in central Europe. One flower-visiting species is *Dasytes plumbeus*. Imagines are black or blue-metallic with erect stiff hairs on and more fine hairs below the elytra. Larvae are predators in decaying wood, whereas imagines are found on shrubs and flowers (Harde and Severa 2006).

#### Curculionidae (true weevils or snout beetles)

Weevils form the superfamily of Curculionoidea (Bouchard et al. 2011), with an estimated number of 220,000 species worldwide, are herbivorous beetles. They are distributed in nearly all latitudes and altitudes with vegetation (McKenna et al. 2009). The Curculionidae are an important family in the superfamily, with appr. 80% of known weevils (McKenna et al. 2009).

In central Europe appr. 1,200 species belonging to 168 genera of true weevils are described (Harde and Severa 2006). One type of weevils, the *Larinus turbinatus*-type, lay their eggs into open flower heads of thistles (Cardueae) and related thistle-like tribes (Zwölfer and Stadler 2004). *L. turbinatus* is 4 to 9 mm in body length, the body is robust and oval and clothed with pathches of grey setae (Hoebeke and Spichiger 2016). This species feed on leaves, stems, buds, flowers, penduncles and possibly pollen of their host (Hoebeke and Spichiger 2016).

#### Meloidae (blister beetles)

The Meloidea, commonly known as blister beetles, form a family in the superfamily of Tenebrionoidea (Bouchard et al. 2011). There are appr. 120 genera and 3,000 species described worldwide (Bouchard et al. 2017; Sharma and Singh 2018). In central Europe, 12 genera with appr. 37 species are native (Harde and Severa 2006). The imagines feed on leaves, pollen and nectar whereas the larvae are parasitic and prey on brood of e.g. the genera Andrena or Orthoptera (Harde and Severa 2006). A native species in Europe is *Lytta vesicatoria*, the Spanish fly. Adults occur from early May until August and feed on leaves of a range of trees and shrubs, mostly on ash, lilac and privet but also willows, honey suckle, rose and various fruit trees among others<sup>5</sup>.

Wilhelmi and Krenn (2012) give an overview of mouthparts and feeding behaviour of the family Meloidae, concluding that next to some taxa where adults do not feed at all, some subgroups have mouthparts of the biting-chewing type, others mouthparts modified to take up nectar.

### Mordellidae (tumbling flower beetles)

The family Mordellidae (superfamily Tenebrionoidea, see Bouchard et al. 2011) are worldwide distributed and can be found in a variety of ecosystems. So far, 2,308 Mordellidae species belonging to 115 genera have been recorded. Many of them act as pollinators, others are important agricultural and forestry pests (Liu et al. 2018).

Species of this family are usually small and wedge-shaped and covered by silky hairs. Adults are found on dead or partly dead trees, but many frequent flowers. Larvae live in dead trees (Liljeblad 1945).

In central Europe, 101 species in 13 genera are described for the Mordellidae, with *Mordella brachyura* and *Mordellistena brevicauda* two common species (Harde and Severa 2006). Another flower-visiting species is *Cteniopus flavus*, the sulphour beetle.

<sup>&</sup>lt;sup>5</sup> <u>https://www.ukbeetles.co.uk/lytta-vesicatoria</u> (14.04.2021)

### Oedemeridae (false blister beetles, pollen-feeding beetles)

The false blister beetles are a family in the superfamily Tenebrionoidea (Bouchard et al. 2011). In central Europe, there are 32 species belonging to 10 genera. Larvae develop in decaying wood or in dry herb stems. Imagines can be found on flowers, foraging pollen (Harde and Severa 2006). According to Sivilov et al. (2011) and Peris (2017) most (if not all) adults feed on pollen and nectar of different plant species, while larvae feed on decaying wood. One prominent and common species in Europe is *Oedemera nobilis*, commonly known as the false oil beetle or the thick-legged flower beetle. This species was, next to *Brassicogethes aeneus*, found with high dominance (depending on the portion of surrounding arable land) on isolated wildflower plantings by Grass et al. (2016) in central Germany. Six different species of *Oedemeridae* could be found on grassland, visiting flowers, in Germany (Weiner 2016), the most numerous being *Oedemera lurida* and *O. femorata. Oedemera femorata* is one of the most common European beetle species found on flowers. Imagines are 8 to 10 mm long and they prefer white blossoms to forage for pollen (Bílý 1990).

### Erotylidae (pleasing fungus beetles)

The family Erotylidae of the superfamily Cucujoidea (Bouchard et al. 2011) is distributed worldwide and contains phytophagous, mycophagous and saprophagous species and some other species that feed on pollen and in dead wood (Leschen and Buckley 2007).

### **Infraorder Elateriformia**

### Buprestidae (jewel beetles)

The family Buprestidae is classified by Bouchard et al. (2011) in the superfamily Buprestoidea. 123 species of 25 genera are common in central Europe (Harde and Severa 2006). Larvae of the family Buprestidae develop mostly in wood or plant stems, whereas imagines can be found on their host plants and on yellow flowers (Harde and Severa 2006). Weiner (2016) reported two species (*Agrilus biguttatus* and *Anthaxia quadripunctata*) on flowers in grassland in Germany. Adults of other species feed on needles and twigs of healthy trees (e.g. *Melanophila* spp. and *Phaenops* spp.) or on fungi (some members of the genus Agrilus) (Bright 1987; Bellamy and Nelson 2002; both cited in O'Neill et al. 2008).

### Elateridae (click beetles)

The Elateridae, or click beetles, are a family in the superfamily Elateroidea (Bouchard et al. 2011) and there are appr. 400 genera with 10,000 species worldwide (Bouchard et al. 2017). In central Europe, appr. 172 species belonging to 52 genera are described (Harde and Severa 2006). They are mostly known as agricultural pests (Bouchard et al. 2017), larvae e.g. feeding on plant roots (Harde and Severa 2006), others are beneficial predators, e.g. of wood-boring beetles or defoliators (Bouchard et al. 2017). A species, where the imagines are known to frequent the flowers of Apiaceae, is *Ampedus sanguineus*.

### Cantharidae (soldier beetles)

The family Cantharidae, commonly known as soldier beetles, are classified by Bouchard et al. (2011) in the superfamily Elateroidea. In central Europe, 104 species belonging to 9 genera are native and the imagines can be found in the summer, often in great abundance, on shrubs and flowers (Harde and Severa 2006). Weiner (2016) collected 7 different species on grassland flowers, *Cantharis fusca*, *Rhagonycha fulva and R. nigriventris* being the most numerous ones. Adults can be found on foliage and on flowers, where they feed on small insects, nectar and pollen (Pelletier and Hébert 2014).

### Infraorder Staphyliniformia

### Hydrophilidae (water scavenger beetles)

The family *Hydrophilidae* is part of the superfamily Hydrophiloidea (Bouchard et al. 2011). One third of the species in this family is known to be secondarily terrestrial (Minoshima et al. 2018). One example researched by Minoshima et al. (2018) is the genus *Rygmodus*, where adults are flower-visiting and pollen-feeding, whereas larvae are aquatic predators.

### Staphylinidae (rove beetles)

The rove beetles are part of the superfamily Staphylinoidea (Bouchrad et al. 2011) and are a diverse group in regard to their feeding behaviour and inhabited microhabitats. Some species are predators, some fungivores, detrivores or pollen-feeders (e.g. some species of the subfamily Omaliinae) (Klimaszewski et al. 2013).

### 3.4.2 Habitat types

Beetles are common in most terrestrial and freshwater habitats (Table 11) and some marine environments (see Bouchard et al. 2017 and references therein) and became adapted to different ecosystems, e.g. the families Dytiscidae and Hydrophilidae live for most of their life-cycle in water (Harde and Severa 2006). Bílý (1990) summarizes, that beetles live in all regions except the polar regions and on ice-covered mountain tops.

Based on the biotope or ecosystem they inhabit, specialised morphological adaptions have developed, e.g. rowing legs within the water beetles (Bílý 1990).

Habitat type	Life stages	Function
Flowers	Adults	Food source (pollen, nectar), nesting, prey hunting
Other plant structures (e.g. leaves, stems, twigs)	Larvae, adults	Food source, nesting, shelter, prey hunting
Soil	Larvae, adults	Nesting, prey hunting, food source (e.g. on plant roots, subterrean fungi)
Water	Larvae, Adults	Food source, water consumption, nesting, shelter
Other organic matter (e.g. litter, animal carcasses)	Larvae, Adults	Food source

**Table 11:** Typical habitat types of Coleoptera in different life stages and their function.

### 3.4.3 Ecological role

In general, beetles play important roles in ecosystems by their diverse interactions with plants and other organisms and with dead or decaying biotic materials (McKenna 2018). Many beetles are beneficial (e.g. nutrient-recyclers, pollinators), others are economically important pests of crops or stored products (Wielkopolan and Obrepalska-Steplowska 2016).

There are many species where larvae are xylophagous, therefore living in rotten or decaying wood, whereas the imagines forage on flowers and pollen (anthophagous or palynophagous). Examples are species from the families Alleculinae, Mordellidae, Oedemeridae, Cerambycidae and Buprestidae (Bílý 1990).

The Coleoptera are a group of insects associated with pollination, including distinct lineages from broadly generalistic to extremely specialized taxa (Parizotto and Grossi 2019). Kevan and Baker (1983) state Elateridae, Scarabeidae, Cleridae, Nitidulidae, Chrysomelidae, Staphylinidae, Meloidae, and Cerambycidae as notable flower visitors in the suborder Polyphaga. According to the literature review by Rader et al. (2020), of the 105 crops that benefit from pollination 51% were visited by Coleoptera, e.g. by members of the families Coccinellidae and Nitidulidae.

According to Bernhardt (2000), there are more than 184 angiosperm species subdivided into 85 genera and representing 34 families that are pollinated almost exclusively by beetles. Representatives from 14 beetle families are associated with these flowers, most of the presented plant genera are visited partly, or exclusively, by members of the family *Scarabaeidae*. In addition, over 98 plant species in 40 genera representing 22 families are pollinated by a combination of beetles and other animals. While the majority of species are represented by magnoliids and basal monocotyledons, specialized beetle-pollinated systems have evolved independently in 14 families of eudicotyledons and six families of petaloid monocots. Also, Bernhardt (2000) concludes, that beetle-pollinated plant species remain understudied since many cantharophilic plants are tall forest trees and many pollinating beetles are nocturnal.

For Europe, the results of Grass et al. (2016) highlight the role of beetles as flower visitors. The authors studied the abundance of flower-visitors on wildflower plantings in Germany. Next to the prominent flower visitors (e.g. *A. mellifera*, wild bees and hoverflies) non-prominent flower visitors could be found on the flowers, of which appr. 15% were Coleoptera. A report of the German Federal Environment Agency (UBA TEXTE 54/2019 and reference within) summarizes the results of the biodiversity explanatories. In this project, the flower-visiting community of three different regions in Germany were examined. The results show that flies were the most abundant group in species and individual observations. The abundancies of bees and beetles were comparable in relation to species and individual observations. In contrast to most bee species, many NBP (e.g. flies, beetles) are capable of larger flight distances (Rader et al. 2020).

### 3.4.4 Feeding behaviour

About 35% of the Coleoptera are herbivorous (Schoonhoven et al. 1998, cited in Romeis et al. 2005), both adults and larvae feed on different plant parts (Romeis et al. 2005). Direct nectar-feeding by beetles seems rare, although flower-visiting species are known to ingest some nectar (Samuelson et al. 1994, cited in Romeis et al. 2005). As summarized in Table 12, there are many species known to feed on pollen and some of them also to contribute to pollination. Based on this overview, the family Oedemeridae seems to be the only family which solely feeds on pollen, although the data reviewed here are not extensive.

	Scar	Cocc	Niti	Phal	Cera	Chry	Cler	Mely	Curc	Melo	Mord	Ored	Bupr	Elat	Cant
A	No	No <sup>6</sup>	No	-	No	Yes	No	No	No						
1	No	No	_	_	No	_									
L							110	110	110		110			110	

(A=adults, L=larvae, Scarabaeidae=Scar, Coccinelidae=Cocc, Nitidulidae=Niti, Phalacridae=Phal, Cerambycidae=Cera, Chrysomelidae=Chry, Cleridaee=Cler, Melyridae=Mely, Curculionidae=Curc, Meloidae=Melo, Mordellidae=Mord, Oredemeridae=Ored, Buprestidae=Bupr, Elateridae=Elat, and Cantharidae=Cant).

<sup>&</sup>lt;sup>6</sup> mostly not, sometimes commitment their diet with pollen

# 4. Sensitivity of non-bee pollinators

## 4.1 Literature review

As it has been described in a scoping paper (ECHA 2020), there is much less data available for non-bee species in order to have a complete understanding of species sensitivity and comparing the results to that of the HB.

Some studies have been previously performed with the objective of comparing the sensitivity of other species to that of the HB. For example, Hardstone and Scott (2010) performed an extensive literature review to compare the toxicity data for HBs and other insect species.

Sensitivity NBP compared to HBs seems to be dependent on the chemical class of the active substance studied (Arena & Sgolastra 2014; Hardstone & Scott 2010; Uhl et al. 2019). In these publications it was found that, for example, for pyrethroids HBs are most sensitive while for neonicotinoids other bees and insect species are more sensitive than HBs. A more extensive summary of sensitivity data for different bee species can e.g., be found in EFSA (2012).

Below, we collected available literature data on the sensitivity of Diptera, Lepidoptera, non-bee Hymenoptera (Symphyta), and Coleoptera to insecticides. The aim was to establish, whether their sensitivity significantly differs to that of the HB, and thus would need to be considered in the environmental risk assessment. Theoretically, it could be achieved either by testing representative non-bee arthropod species or by appropriate assessment factors.

#### 4.1.1 Order Diptera

Databases used were Pubmed, Science Direct, and Researchgate.net.

The search included the following families of Diptera: Muscidae, Nemestrinidae, Stratiomyidae, Tabanidae, Conopidae, Calliphoridae, Bombyliidae and Syrphidae. Key words and their combinations used were as follows:

"Muscidae" OR "Nemestrinidae" OR "Stratiomyidae" OR "Tabanidae" OR "Conopidae" OR "Calliphoridae OR "Bombyliidae" OR "Syrphidae OR "Diptera" AND "insecticides" OR "pesticides" OR "LD50" OR "EC50" OR "toxicity" OR "biocides".

A total of 12 studies were selected as relevant and further evaluated (see Annex I).

The most commonly used species in laboratory studies is *Musca domestica* (family Muscidae), either larvae stages or adults. This species is worldwide common and used regularly for ecotoxicological analysis. Also, other species from the family *Muscidae* were used as test species in the studies, such as *Haematobia irritans*, *Musca automnalis* (species which occurs mostly in stables), but also species from other families like *Eristalis tenax* (Family Syrphidae). The other used species of the order Diptera, *Culex pipiens* (Family Culicidae) and *Fannia canicularis* (Family Fanniidae) are not in the scope of the related dipteran families. In many studies approved biocidal active substances for PT18 and PT19 under the BPR were used as test substances<sup>7</sup>:

<sup>&</sup>lt;sup>7</sup> <u>https://echa.europa.eu/de/regulations/biocidal-products-regulation/approval-of-active-substances/list-of-approved-active-substances</u>

PT18: Cypermethrin, Diflubenzuran, Permethrin, Piperonyl butoxide, Spinosad, Thiamethoxam

### PT19: N-Diethyl-Meta-Toluamide (DEET)

However, the studies cannot be considered representative for the assessment of sensitivity compared to HBs for the following reasons:

Although in most of the studies PT18 substances were used and for those cases, agreed bee data in the biocide and PPP regime is available, the results for  $LD_{50}/EC_{50}/LC_{50}$  are expressed in units such as  $\mu g/vial$ ,  $\mu g/cm^2$ , ppb, mM, mg/m<sup>2</sup> or mg/kg. Hence, such results cannot be compared directly to bee data, which are expressed for acute tests as  $\mu g$  test substance/bee.

In the study of Meyer et al. (1990) the LD<sub>50</sub> (contact, topical application of permethrin) for *Fannia canicularis* is expressed in the same unit as for acute bee tox tests: LD<sub>50</sub> =  $0.022 \mu g/fly$ . Furthermore, the same publication gives an LD<sub>50</sub> =  $0.0233 \mu g/fly$  for *Musca domestica* when exposed to pyrethrins/PBO (mixture 5:1).

However, the duration of the study and further methological aspects (e.g. concentrations used, details on the statistical analysis) were not stated in the study report. Nevertheless, the value was used when comparing the acute contact toxicity endpoints for different species exposed to permethrin (see Table 21).

Sukontason et al. (2005) performed bioassays with Musca domestica (house fly) and *Chrysomya megacephala* (blow fly). Both species were tested according to WHO guideline (1980): 1  $\mu$ l of the test substances permethrin and deltamethrin were topically applied to anesthetized adult flies. The 24h-LD<sub>50</sub> values for the laboratory strain of *M. domestica* are 0.0049  $\mu$ g/fly (permethrin) and 0.158  $\mu$ g/fly (deltamethrin). For the laboratory strain of *C. megacephala* the LD<sub>50</sub> are 0.0028  $\mu$ g/fly (permethrin) and 0.0461  $\mu$ g/fly (deltamethin).

Mostly, *Musca domestica* was used as test species, which is only one species representing the order Diptera (family Muscidae). Since the adults of *Musca domestica* feed also on sugar-containing fluids, nectar can be a food source. Only for the representative family Calliphoridae one relevant data point was found. Since in Europe about 45 genera and 575 species (Oosterbroek 2006) are described, it is highly questionable if the test species *Musca domestica* can represent the whole diversity of the Muscid family or even the order Diptera. Due to the small number of reliable studies, it was decided not to search for comparable toxicity studies with HBs in the open literature.

Furthermore, most of the studies dealt with resistance and are therefore not comparable with standard bee toxicity tests.

It can be concluded, that due to the small number of reliable studies available (which were in fact only two) their relevance to the question of sensitivity for the order Diptera compared to HBs cannot be answered at present. Further research is needed for studies with different species of flower-visting dipteran species to gain a better understanding of the issue of sensitivity of Diptera compared to HBs.

### 4.1.2 Order Lepidoptera

Lepidoptera (butterflies and moths) are an important group for which standard test protocols are missing. For larvae, there are test guidelines that could be adapted for oral and contact exposure (Lang et al. 2019). Furthermore, an oral toxicity study with lepidopteran larvae representatives of herbivorous species is recommended by EFSA (2015), and further test methods that would include effects from chronic exposure and delayed effects in non-target arthropods in the lower tiers will be developed. This should allow for estimating effects on the most crucial life history parameters, such as longevity and reproduction rate (UBA-Texte 54/2019).

A literature search was conducted on the chosen Lepidoptera species (see Table 2) to evaluate if they are more sensitive to biocides than HBs. The keywords and combinations used in the search were as follows:

"Lepidoptera" OR "Sphingidae" OR "Zygaenidae" OR "Hesperiidae" OR "Papilionidae" OR "Nymphilidae" OR "Pieridae" OR "Lycaenidae" AND "toxicity" OR "biocides" OR "LD50" OR "sensitivity"

Overall, 19 publications were found (see Annex II). Based on the literature review, butterflies and moths can be considered as sensitive species but it is difficult to compare the data to that of the HB. In the case of butterflies, this is due to the large amount of extremely varying data: the studies have been done at a variety of species, different life forms, and different exposure ways (thorax, wings, contact, leaf-dip, etc.), and additionally, the reporting of the results varies:  $LD_{50}/LC_{50}$  can be provided as  $\mu g/g$ , ppm, mg/L; mg/cm<sup>2</sup> or the toxicity has been reported as  $LD_{10}/LD_{90}/LC_{20}$  etc. For moths, on the other hand, there are not many published studies for their sensitivity. The overall variability of the data makes a sensitivity analysis on Lepidoptera extremely difficult with the available resources.

Some of these studies have included a comparison to HB sensitivity. Lepidoptera have been suggested to be even more sensitive than HBs for certain substances, such as naled and permethrin (Hoang et al. 2011; Hoang et al. 2015), but the small sample size leaves room for uncertainty. For example, Hoang et al. (2011) compared the LD<sub>50</sub> of the butterflies from their study with HB LD<sub>50</sub> and concluded that several butterfly species are more sensitive to these insecticides than HBs. They further stated that HB is not a good representative of insects, as they have a large surface area per volume ratio (such as butterflies) and which are more likely to encounter higher exposure concentrations in the field.

Nymphalidae, Lycaenidae, Hesperiidae and Papilionidae are the well studied Lepidoptera families. However, despite the wide use of insecticides and the acknowledged importance of Lepidoptera, it has yet been impossible to determine which species is the most sensitive or which insecticide is the most toxic to the species, because of the small number of published studies, different methodological approaches, and differently expressed endpoints (Mulé et al. 2017). Two species that have been investigated in several of the collected studies are *Pieris brassicae* and *Pieris rapae*.

### 4.1.3 Order Hymenoptera

### **Suborder Apocrita**

A literature review was conducted to study the sensitivity of the chosen wasp species (see chapter 5.2.2) and evaluate if they are more sensitive to biocides than HBs. The keywords and combinations used in the search were as follows:

"Crabronidae" OR "Sphecidae" OR "Chrysididae" OR "Pompilidae" OR "Tiphiidae" OR "Scoliidae" OR "Vespidae" AND "insecticides" OR "biocides" OR "pesticides" OR "toxicity" OR "LD50" OR "sensitivity"

Only two studies were found using these parameters, as not many studies have been done on wasp sensitivity. There has been some research on comparing wasps to HBs: Fernandes et al. (2008) consider wasps as "unusually sensitive" as HBs to different insecticides. They compared the sensitivity of *P. sylveirae* (Vespidae) and *A. mellifera* to different insecticides and found both species to be equally intolerant to dimethoate, fenpropathrin and thiametoxam, but *P. sylveirae* was more tolerant to triflumuron and spinosad. Yet, there are not many studies available at present on the sensitivity of wasp species to insecticides, among them, Stark et al. (1995) published an LD<sub>50</sub> of 0.0004  $\mu$ g/organism.

Based on the research data available at present, it is considered not possible to go on with a sensitivity analysis to conclude if wasps can be considered more sensitive than the HBs to biocides. The few studies found by the literature review have been presented in the attached Annex III (see chapter 5.2.2). There is a need for more research in this area in order to be able to conclude on the sensitivity of wasps to biocides and other chemicals. For instance, a research report (UBA-Texte 54/2019) has suggested species such as the predatory wasp *Trichogramma cacoeciae* to be tested in addition to HBs. In addition, *Aphidius rhopalosiphi* and *Trichogramma dendrolimi* are considered sensitive indicator species for non-target parasitic wasps.

### Suborder Symphyta

Key words and their combinations used were as follows for Symphyta:

"Coleoptera" OR "Scarabaeidae" OR "Coccinellidae" OR "Nitiludidae" OR "Phalacridae" OR "Cerambycidae" OR "Chrysomelidae" OR "Cleridae" OR "Melyridae" OR "Oedemeridae" OR "Buprestidae" OR "Elateridae" OR "Cantharidae" AND "insecticides" OR "pesticides" OR "LC50" OR "sensitivity" OR "toxicity"

Only one study (Lung 1980) was found providing information about the sensitivity of one species within this group. However, the study was mostly focused on oral exposure of larval stages. Therefore, no comparison with HB sensitivity data is possible. Due to the limited information available and the overwhelming absence of ecotoxicological studies exploring the sensitivity of sawflies to biocides, it is not possible to draw any conclusion about the sensitivity of this group. This lack of information requires a greater effort to advance in the knowledge of this group, thus facilitating the evaluation of its sensitivity to biocides by comparing it with the HB.

### 4.1.4 Order Coleoptera

Key words and combinations used in this search were as follows:

"Coleoptera" OR "Scarabaeidae" OR "Coccinellidae" OR "Nitiludidae" OR "Phalacridae" OR "Cerambycidae" OR "Chrysomelidae" OR "Cleridae" OR "Melyridae" OR "Oedemeridae" OR "Buprestidae" OR "Elateridae" OR "Cantharidae" AND "insecticides" OR "pesticides" OR "LC50" OR "sensitivity" OR "toxicity"

Based on the information available in the abstract, roughly 65 publications were selected and more detailed information on the test (e.g. test species, chemicals used, application method) gathered from the full text. Based on this information, 16 publications were considered useful for a sensitivity comparison with bee toxicity data (Annex IV) as information on dose-response are given. The focus is on studies using topical application of a known concentration on individuals. Effect values were given or recalculated as concentration per individuum. For a direct comparison with bee effect values the mean bodyweight of the beetles (larvae or adults) was included in the table, as far as available.

The 16 selected studies cover a range of biocidal active substances (e.g. imidacloprid, thiamethoxam, acetamiprid, lambda-cyhalothrin, etofenprox, abamectin, permethrin) of different chemical classes, while exposure of test organisms was either oral or by topical application. A range of coleopteran species were used, the majority belonging to the family Coccinellidae.

# 4.2 Comparison of sensitivity data

### **Presentation of data**

As described in the previous paragraphs, a comparison of sensitivity data between bee and nonbee species is only possible for some representatives of the orders Lepidoptera and Coleoptera, and one dipteran species. Data for bee species were collected from open literature, active substance assessment reports (ECHA, EFSA) and/or data bases (e.g. PPDB). Only for acute contact tests by topical application LD<sub>50</sub>s as  $\mu$ g/organism could be found or recalculated. The focus of the sensitivity comparison in this document is on this type of exposure to allow comparison of lethal doses. All in all, 119 datapoints were used for the sensitivity analysis, the distribution of datapoints for the nine selected active substances is shown in Figure 9.

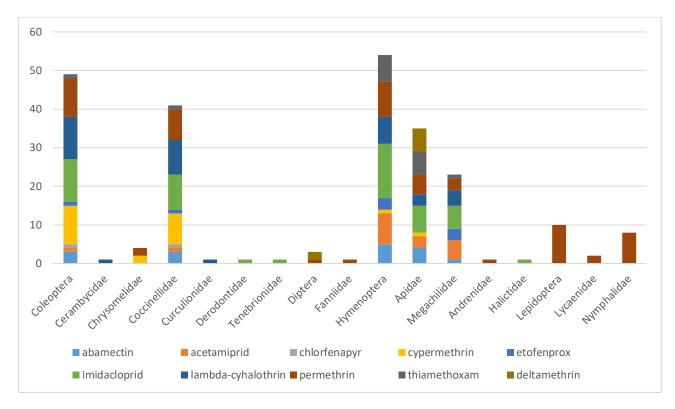


Figure 2: Number of reported datapoints used for comparison of sensitivity.

As individual weight has an influence on sensitivity towards insecticides and is therefore recommended to be reported in addition to the  $LD_{50}$  in µg/organism (Uhl et al. 2019), both the  $LD_{50}$  in µg/organism and the weight-normalised  $LD_{50}$  are presented in the tables in this chapter. A mean body weight was used when more than one reference was available. An overview of endpoints and methological details as well as the references for endpoints and body weight data can be found in the Annex V.

The following pages present endpoints for different species of FVI and 10 active substances. In the tables the most sensitive endpoint for each order is highlighted in bold. Additionally, the most sensitive endpoint for HB is also highlighted in bold, as it is often suggested as a surrogate species.

#### <u>Thiamethoxam</u>

The active substance thiamethoxam, a neonicotinoid insecticide (IRAC sub group 4A<sup>8</sup>), was approved for use in PT18 (Insecticides, acaricides and products to control other arthropods) in 2015<sup>9</sup>.

The data for acute contact tests with different bee species shows that the solitary bee *Osmia lignaria* seems to be more sensitive when exposed to thiamethoxam than *Apis mellifera* (factor of 25 for LD<sub>50</sub> in  $\mu$ g/organism). The endpoints for the most sensitive stingless bee species (*Nannotrigona perilampoides*) are a factor of 6 lower compared to the endpoint for HB. The contact endpoints available for the beetle species *Harmonia axyridis* are by appr. a factor of 6 higher than the enpoints for HB. As an LD<sub>50</sub> is only available for one beetle species, a conclusion on sensitivity for the highly diverse group of NBP cannot be drawn.

Table 13: Comparison of acute toxicity endpoints for different species topically exposed to thiamethoxam

Species tested	Order (family)	Test duration (hours)	LD <sub>50</sub> in µg/organism	Weight normalised LD <sub>50</sub> in µg/g organism	Reference
<i>Harmonia axyridis</i> (harlequin ladybird beetle)	Coleoptera (Coccinellidae)	48	0.151	4.03	Youn et al. (2003)
<i>Bombus terrestris</i> (buff-tailed bumble bee)	Hymenoptera (Apidae)	n.a.	0.028	0.13	PPDB (verified data used for regulatory purposes)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	48	0.024	0.2	Assessment Report PT 18 (ECHA, 2012); EFSA Journal 2013
<i>Melipona quadrifasciata</i> (stingless bee) <sup>10</sup>	Hymenoptera (Apidae)	48	0.0091	1.12	Piovesan et al. (2020)
<i>Tetragonisca fiebrigi</i> (stingless bee) <sup>10</sup>	Hymenoptera (Apidae)	48	0.0055	n.a.	Piovesan et al. (2020)

<sup>&</sup>lt;sup>8</sup> <u>https://irac-online.org/modes-of-action/</u>

<sup>&</sup>lt;sup>9</sup> <u>https://echa.europa.eu/information-on-chemicals/biocidal-active-substances</u>

<sup>&</sup>lt;sup>10</sup> eusocial, native to South America

<i>Trigona iridipennis</i> (dammar bee) <sup>11</sup>	Hymenoptera (Apidae)	n.a. <sup>12</sup>	0.0051	0.51	Kumar and Regupathy (2005) <sup>13</sup> (cited in the PPDB)
<i>Nannotrigona perilampoides</i> (Mexican pitted stingless bee) <sup>14</sup>	Hymenoptera (Apidae)	24	0.004	0.55	Valdovinos- Núñez et al. (2009)
Osmia lignaria (orchard mason bee) <sup>15</sup>	Hymenoptera (Megachilidae)	96	0.0011	0.0097	Peterson et al. (2021)

n.a. information not available

#### **Imidacloprid**

The active substance imidacloprid, a neonicotinoid insecticide (IRAC subgroup  $4A^{16}$ ), was approved for use in PT18 in 2013<sup>17</sup>.

Table 15 summarizes acute toxicity endpoints for different bee and beetle species and one wasp species. The most sensitive species when exposed to imidacloprid by contact is *Sasajiscymnus tsugae*, a coccinellid beetle. The most sensitive bee species is *Apis mellifera ssp. mellifera*, which is by a factor of appr. 10 less sensitive than *Sasajiscymnus tsugae*, although the exposure time is different (48h-LD<sub>50</sub> for *Apis mellifera* vs. 144h-LD<sub>50</sub> for *Sasajiscymnus tsugae*). Weightnormalised endpoints show, that *Osmia lignaria* is the most sensitive species when topically exposed to imdacloprid.

<sup>14</sup> native to South America

<sup>&</sup>lt;sup>11</sup> tropical stingless bee species, eusocial

<sup>&</sup>lt;sup>12</sup> data not available

<sup>&</sup>lt;sup>13</sup> full text not available

<sup>&</sup>lt;sup>15</sup> native to North America

<sup>&</sup>lt;sup>16</sup> <u>https://irac-online.org/modes-of-action/</u>

<sup>&</sup>lt;sup>17</sup> <u>https://echa.europa.eu/information-on-chemicals/biocidal-active-substances</u>

Table 14: Comparison of acute toxicity endpoints	for different species topically exposed to imidacloprid
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Species tested	Order (family)	Test duration (hours)	LD <sub>50</sub> in µg/organism	Weight normalised LD <sub>50</sub> in µg/g organism	Reference
<i>Osmia</i> <i>cornifrons</i> (Japanese hornfaced bee) <sup>18</sup>	Hymenoptera (Megachilidae)	48	3.82	29.16	Biddinger et al. (2013)
<i>Harmonia axyridis</i> (harlequin ladybird beetle)	Coleoptera (Coccinellidae)	48	0.36	9.6	Youn et al. (2003)
<i>Bombus</i> <i>terrestris</i> (buff-tailed bumble bee)	Hymenoptera (Apidae)	96	0.218	0.99	EFSA Journal (2018)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	48	0.15	1.25	Biddinger et al. (2013)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	48	0.081	0.675	Assessment Report PT 18 (ECHA, 2015)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	24	0.04	0.34	Stark et al. (1995)
Coleomegilla maculate (twelve- spotted ladybird beetle)	Coleoptera (Coccinellidae)	48	0.074	5.21	Lucas et al. (2004)
Osmia bicornis (red mason bee)	Hymenoptera (Megachilidae)	48	0.03	0.33	Uhl et al. (2019)
Scaptotrigona postica (stingless bee) <sup>19</sup>	Hymenoptera (Apidae)	48	0.025	0.824	Soares et al. (2015)
<i>Bombus terrestris</i> (buff-tailed bumble bee)	Hymenoptera (Apidae)	72	0.02	0.09	Marletto et al. (2003)

<sup>18</sup> native to Northern Asia

<sup>19</sup> native species in Brazil, eusocial

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	1		1	T	
<i>Cryptolaemus montrouzieri</i> (mealybug ladybird)	Coleoptera (Coccinellidae)	24	0.0173	2.01	Khani et al. (2012)
<i>Martianus dermestoides</i> (Chinese beetle)	Coleoptera (Tenebrionidae)	142	0.0135	n.a. <sup>20</sup>	Guan et al. (2008)
<i>Apis mellifera ssp. caucasia</i> (caucasian honey bee)	Hymenoptera (Apidae)	48	0.0128	n.a.	Suchail et al. (2000)
<i>Apis mellifera ssp. mellifera</i> (honey bee)	Hymenoptera (Apidae)	48	0.024	0.2	Suchail et al. (2000)
<i>Hippodamia</i> <i>convergens</i> (convergent lady beetle)	Coleoptera (Coccinellidae)	72	0.006	0.4	Kaakeh et al. (1996)
Hippodamia convergens (convergent lady beetle)	Coleoptera (Coccinelidae)	24	0.010	0.68	Stark et al. (1995)
<i>Osmia lignaria</i> (orchard mason bee) <sup>21</sup>	Hymenoptera (Megachilidae)	96	0.0028	0.0255	Peterson et al. (2021)
<i>Laricobius nigrinus</i> (hemlock woolly adelgid)	Coleoptera (Derodontidae)	144	0.0018	2.4	Eisenback et al. (2010)
<i>Nannotrigona</i> perilampoides (stingless bee) 22	Hymenoptera (Apidae)	24	0.0011	0.15	Valdovinos- Núñez et al. (2009)
<i>Sasajiscymnus tsugae</i> (ladybird beetle) <sup>23</sup>	Coleoptera (Coccinellidae)	144	0.00071	1.82	Eisenback et al. (2010)
<i>Megachile rotundata</i> (alfalfa leafcutting bee)	Hymenoptera (Apidae)	n.a.	0.17	5.79	PPDB

<sup>20</sup> data not available

<sup>21</sup> native to North America
 <sup>22</sup> neotropical species

<sup>23</sup> native to Japan

<i>Megachile rotundata</i> (alfalfa leafcutting bee)	Hymenoptera (Apidae)	24	0.04	1.39	Stark et al. (1995)
<i>Nomia melanderi</i> (alkali bee)	Hymenoptera (Apidae)	24	0.04	0.46	Stark et al. (1995)
<i>Aphidius ervi</i> (braconid wasp)	Hymenoptera (Braconidae)	24	0.0004	0.76	Stark et al. (1995)

### <u>Acetamiprid</u>

The active substance acetamiprid, a neonicotinoid insecticide (IRAC subgroup  $4A^{24}$ ), was approved for use in PT18 in  $2020^{25}$ .

Acetamiprid is less acute toxic to the tested bee species (see Table 16). The most sensitive endpoint published by Youn et al. (2003) was found for the coccinellid beetle *Harmonia axyridis*. Thus, this species is by a factor of ~100 more sensitive than *Osmia bicornis* and by a factor of ~470 more sensitive than *Apis mellifera*. Even when weight-normalising the endpoints, the harlequin ladybird beetle is by a factor of 40 more sensitive than the most sensitive bee species (*Osmia bicornis*) and by a factor of 150 more sensitive than *Apis mellifera*.

<sup>&</sup>lt;sup>24</sup> <u>https://irac-online.org/modes-of-action/</u>

<sup>&</sup>lt;sup>25</sup> <u>https://echa.europa.eu/information-on-chemicals/biocidal-active-substances</u>

Table 15: Comparison of	acute toxicity endpoints for	different species topically exp	osed to acetamiprid.
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Species tested	Order (family)	Test duration (hours)	LD <sub>50</sub> in µg/organism	Weight normalised LD₅₀ in µg/g organism	Reference
<i>Bombus terrestris</i> (buff-tailed bumble bee)	Hymenoptera (Apidae)	48	>100	>451.47	PPDB; Sanchez- Bayo and Goka (2014) (table S2) (unverified data)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	48	64.6	538.33	Biddinger et al. (2013)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	48	9.26	77.17	Assessment Report PT 18 (ECHA, 2018)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	72	8.1	67.5	PPDB (verified data used for regulatory purposes)
<i>Plebeia emerina</i> (stingless bee) <sup>26</sup>	Hymenoptera (Apidae)	48	6.22	n.a.	Padilha et al. (2020)
<i>Osmia</i> <i>cornifrons</i> (Japanese hornfaced bee) <sup>27</sup>	Hymenoptera (Megachilidae)	48	4.0	30.53	Biddinger et al. (2013)
<i>Teragonisca fiebriegi</i> (stingless bee) <sup>28</sup>	Hymenoptera (Apidae)	48	1.42	n.a.	Padilha et al. (2020)
<i>Osmia bicornis</i> (red mason bee)	Hymenoptera (Megachilidae)	48	1.72	18.11	Uhl et al. (2019)
<i>Harmonia axyridis</i> (harlequin ladybird beetle)	Coleoptera (Coccinellidae)	48	0.017	0.45	Youn et al. (2003)

<sup>26</sup> native to South America
<sup>27</sup> native to Eastern Asia

<sup>&</sup>lt;sup>28</sup> eusocial, native to South America

#### lambda-Cyhalothrin

The active substance lambda-cyhalothrin, a pyrethroid insecticide (IRAC subgroup  $3A^{29}$ ), was approved for use in PT18 in 2013<sup>30</sup>.

For lambda-cyhalothin the most sensitive acute endpoint (contact) is available for *Curinus coeruleus*, the metallic-blue lady beetle. Compared to *Apis mellifera*, this beetle species is appr. 100 times more sensitive when topically exposed. Sensitivity between different beetle species tested by Rodrigues et al. (2013a and 2013b) show a high diversity between endpoints, in fact between the most sensitive ( $LD_{50} = 0.00035 \mu g$ /beetle for *Curinus coeruleus*) and the least sensitive ( $LD_{50} = 0.02 \mu g$ /beetle for *Eriopis connexa*) is a factor of 57, if only focusing on the species tested in Rodrigues et al. (2013a). When weight-normalising the  $LD_{50}$ s, the HB endpoint would be in the same range as the lowest endpoints for Coleoptera species.

**Table 16:** Comparison of acute toxicity endpoints for different species topically exposed to lambdacyhalothrin.

Species tested	Order (family)	Test duration (hours)	LD₅₀ in µg/orga nism	Weight normalised LD <sub>50</sub> in µg/g organism	Reference
<i>Osmia</i> <i>cornifrons</i> (Japanese hornfaced bee) <sup>31</sup>	Hymenoptera (Megachilidae)	48	0.91	6.95	Biddinger et al. (2013)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	48	0.3	2.5	Biddinger et al. (2013)
<i>Osmia bicornis</i> (red mason bee)	Hymenoptera (Megachilidae)	48	0.14	1.45	Uhl et al. (2019)
Anoplophora glabripennis (Asian long- horned beetle)	Coleoptera (Cerambycidae)	24	0.136	1.13	Wu and Smith (2015)
<i>Bombus</i> <i>terrestris</i> (buff- tailed bumble bee)	Hymenoptera (Apidae)	72	0.11	0.50	Marletto et al. (2003)
Hippodamia convergens (convergent ladybird beetle)	Coleoptera (Coccinellidae)	24	0.068	4.55	Rodrigues et al. (2013a)

<sup>&</sup>lt;sup>29</sup> <u>https://irac-online.org/modes-of-action/</u>

<sup>&</sup>lt;sup>30</sup> <u>https://echa.europa.eu/information-on-chemicals/biocidal-active-substances</u>

<sup>&</sup>lt;sup>31</sup> native to Eastern Asia

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<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	n.a.	0.038	0.32	Assessment Report PT 18 (ECHA, 2011)
<i>Olla v-nigrum</i> (ashy-gray lady beetle) <sup>32</sup>	Coleoptera (Coccinellidae)	24	0.027	n.a.	Rodrigues et al. (2013a)
<i>Cycloneda</i> <i>sanguinea</i> (ladybird beetle) <sup>33</sup>	Coleoptera (Coccinellidae)	24	0.024	1.70	Rodrigues et al. (2013a)
<i>Eriopis connexa</i> (ladybird beetle) <sup>34</sup>	Coleoptera (Coccinellidae)	24	0.02	2.7	Rodrigues et al. (2013a)
<i>Coleomegilla maculata</i> (twelve-spotted lady beetle)	Coleoptera (Coccinellidae)	24	0.007	0.50	Rodrigues et al. (2013a)
<i>Eriopis connexa</i> (ladybird beetle) <sup>34</sup>	Coleoptera (Coccinellidae)	24	0.005	0.68	Rodrigues et al. (2013b)
<i>Hippodamia</i> <i>convergens</i> (convergent lady beetle)	Coleoptera (Coccinellidae)	24	0.005	0.334	Barbosa et al. (2016)
<i>Brumoides foudrasi</i> (ladybird beetle)	Coleoptera (Coccinellidae)	24	0.0045	n.a.	Rodrigues et al. (2013a)
<i>Listronotus maculicollis</i> (annual bluegrass weevil) <sup>35</sup>	Coleoptera (Curculionidae)	24	0.00052	n.a.	Ramoutar et al. (2009)
<i>Curinus coeruleus</i> (dark blue lady beetle) <sup>36</sup>	Coleoptera (Coccinellidae)	24	0.00035	n.a. <sup>37</sup>	Rodrigues et al. (2013a)

<sup>&</sup>lt;sup>32</sup> native to Central America, North America and Oceania

<sup>&</sup>lt;sup>33</sup> not native to Europe, distributed among Latin America and on the Galapagos Islands

<sup>&</sup>lt;sup>34</sup> neotropical distribution

<sup>&</sup>lt;sup>35</sup> native to parts of the USA

<sup>&</sup>lt;sup>36</sup> not native to Europe

<sup>&</sup>lt;sup>37</sup> data not available

#### **Etofenprox**

The active substance etofenprox, a pyrethroid insecticide (IRAC subgroup  $3A^{38}$ ), was approved for use in PT08 in 2010 and in PT18 in  $2015^{39}$ .

**Table 17:** Comparison of acute toxicity endpoints for different species topically exposed to etofenprox.

Species tested	Order (family)	Test duration (hours)	LD₅₀ in µg/organism	Weight normalised LD <sub>50</sub> in µg/g organism	Reference
<i>Harmonia axyridis</i> (harlequin ladybird beetle)	Coleoptera (Coccinellidae)	48	0.263	7.01	Youn et al. (2003)
<i>Osmia bicornis</i> (red mason bee)	Hymenoptera (Megachilidae)	48	0.18	2.09	Uhl et al. (2019)
Megachile rotundata (alfalfa leafcutting bee)	Hymenoptera (Megachilidae)	24	0.051	1.738	Piccolomini et al. (2018)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	n.a. <sup>40</sup>	>0.038	>0.3167	PPBD (verified data used for regulatory purposes)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	72	0.0145	0.12	Assessment Report PT 18 (ECHA, 2013)

The sensitivity of *Megachile rotundata* is in the same range with an LD<sub>50</sub> value by a factor of 5 lower compared to the LD<sub>50</sub> of *Harmonia axyridis*, whereas *Apis mellifera* is by a factor of appr. 18 more sensitive. Based on the weight-normalised LD<sub>50</sub>s, *Apis mellifera* seems to be more sensitive to etofenprox than *Osmia bicornis* and the ladybird beetle species *Harmonia axyridis*.

<sup>&</sup>lt;sup>38</sup> <u>https://irac-online.org/modes-of-action/</u>

<sup>&</sup>lt;sup>39</sup> https://echa.europa.eu/information-on-chemicals/biocidal-active-substances

<sup>&</sup>lt;sup>40</sup> data not available

### <u>Abamectin</u>

The active substance abamectin, an avermectin insecticide (IRAC main group  $6^{41}$ ), was approved for use in PT18 in  $2013^{42}$ .

The most sensitive acute contact  $LD_{50}$  was found for *Apis mellifera* (ECHA, 2011), which is in the same range as the 48h-LD<sub>50</sub> for *Harmonia axyridis*, the convergent lady beetle (Youn et al. 2003). When comparing the weight-normalised  $LD_{50}$ s, the HB seems to be most sensitive when topically exposed to abamectin.

**Table 18:** Comparison of acute toxicity endpoints for different species topically exposed to abamectin.

Species tested	Order (family)	Test duration (hours)	LD₅0 in µg/organism	Weight normalised LD <sub>50</sub> in µg/g organism	Reference
<i>Melipona quadrifasci ata</i> (stingless bee) <sup>43</sup>	Hymenoptera (Apidae)	24	134.6	16,617.28	del Sarto et al. (2014)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	24	7.8	65	del Sarto et al. (2014)
<i>Melipona quadrifasci ata</i> (stingless bee) <sup>43</sup>	Hymenoptera (Apidae)	48	0.24	29.63	Piovesan et al. (2020)
<i>Bombus</i> <i>terrestris</i> (buff-tailed bumble bee)	Hymenoptera (Apidae)	72	0.14	0.63	Marletto et al. (2003)
Cryptolaem us montrouzie ri (mealybug ladybird)	Coleoptera (Coccinellidae)	24	0.06673	7.759	Khani et al. (2012)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	n.a.	0.03	0.25	Sanchez-Bayo and Goka (2014) (table S2)

<sup>&</sup>lt;sup>41</sup> <u>https://irac-online.org/modes-of-action/</u>

<sup>&</sup>lt;sup>42</sup> https://echa.europa.eu/information-on-chemicals/biocidal-active-substances

<sup>&</sup>lt;sup>43</sup> not native to Europe, eusocial

<i>Tetragonisc a fiebrigi</i> (stingless bee) <sup>43</sup>	Hymenoptera (Apidae)	48	0.008	n.a. <sup>44</sup>	Piovesan et al. (2020)
<i>Harmonia axyridis</i> (harlequin ladybird)	Coleoptera (Coccinellidae)	48	0.005	0.877	Youn et al. (2003)
<i>Osmia lignaria</i> (orchard mason bee) <sup>45</sup>	Hymenoptera (Megachilidae)	96	0.0036	0.030	Peterson et al. (2021)
Apis mellifera (honey bee)	Hymenoptera (Apidae)	24	0.0022	0.018	Assessment Report PT 18 (ECHA, 2011)

n.a. information not available

#### <u>Chlorfenapyr</u>

The active substance chlorfenapyr, a pyrrole insecticide (IRAC main group 13<sup>46</sup>), was approved for use in PT08 in 2015 (initial application for approval PT18 still in progress)<sup>47</sup>.

The endpoints available for *Apis mellifera* are in the same range as the LD<sub>50</sub> for *Harmonia axyridis*.

<sup>&</sup>lt;sup>44</sup> Information not available

<sup>&</sup>lt;sup>45</sup> native to North America

<sup>&</sup>lt;sup>46</sup> <u>https://irac-online.org/modes-of-action/</u>

<sup>&</sup>lt;sup>47</sup> https://echa.europa.eu/information-on-chemicals/biocidal-active-substances

Species **Order (family)** Test LD<sub>50</sub> in Weight Reference tested duration µg/organis normalised (hours) LD<sub>50</sub> in m µg/g organism Apis Hymenoptera n.a. 0.023 0.192 PPDB mellifera (Apidae) (honey bee) PPDB Bombus Hymenoptera n.a. 0.119 0.537 (Apidae) terrestris (buff-tailed bumble bee) 5.0 PPDB Trigona Hymenoptera n.a. 0.07 spinipes (Apidae) (spinylegged stingless bee) Coccinella Coleoptera 24 0.029 0.84 Coats et al. transversogu (Coccinellidae) 1979 ttata (n.a.) Coleomegilla Coleoptera 24 0.0007 0.048 Coats et al. maculata (Coccinellidae) 1979 spp. lengi (threebanded lady beetle) 0.0005 Hippodamia Coleoptera 24 0.042 Coats et al. convergens (Coccinellidae) 1979 (convergent lady beetle) Hippodamia Coleoptera 24 0.0165 0.75 Coats et al. glacialis (Coccinellidae) 1979 (glacial lady beetle) 24 0.00075 Hippodamia Coleoptera 0.092 Coats et al. 1979 parenthesis (Coccinellidae) (parenthesis lady beetle) 24 0.0008 0.072 Hippodamia Coleoptera Coats et al. 1979 tredecimpun (Coccinellidae) ctata tibialis (thirteenspot ted lady beetle)

Table 19: Comparison of acute toxicity endpoints for different species topically exposed to chlorfenapyr.

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<i>Diabrotica longicornis</i> (northern corn rootworm)	Coleoptera (Chrysomelidae)	24	0.0012	0.17	Coats et al. 1979
<i>Oulema melanopus</i> (cereal leaf beetle)	Coleoptera (Chrysomelidae)	24	0.002	0.28	Coats et al. 1979
<i>Adalia bipunctata</i> (two-spotted ladybird beetle)	Coleoptera (Coccinellidae)	24	0.00003	0.0026	Coats et al. 1979

#### **Cypermethrin**

The active substance cypermethrin, a pyrethroid insecticide (IRAC sub group 3A)<sup>48</sup> was approved for use in PT 08 in 2015 and in PT 18 in  $2020^{49}$ .

The most sensitive acute contact endpoint is an LD<sub>50</sub> of 0.00003  $\mu$ g/organism for *Adalia bipunctata* (Coats et al. 1979). In comparison, the endpoint for *Apis mellifera*, extracted from the PPDB, is an LD<sub>50</sub> of 0.023  $\mu$ g/organism. For cypermethrin, the most sensitive tested coccinellid beetle species is by a factor of appr. 750 more sensitive than HB.

**Table 20:** Comparison of acute toxicity endpoints for different species topically exposed to cypermethrin.

Species tested	Order (family)	Test duration (hours)	LD <sub>50</sub> in µg/organis m	Weight normalised LD <sub>50</sub> in µg/g organism	Reference
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	96	0.33	2.75	Assessment Report PT 08 (ECHA, 2012)
<i>Harmonia axyridis</i> (harlequin ladybird beetle)	Coleoptera (Coccinellidae)	48	0.21	5.6	Youn et al. (2003)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	n.a. <sup>48</sup>	0.15	1.25	Sanchez- Bayo and Goka (2014) (table S2)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	n.a.	0.12	1.0	PPBD (U.S. EPA ECOTOX unverified)

<sup>48</sup> Information not available

### <u>Deltamethrin</u>

The active substance deltamethrin, a pyrethroid insecticide (IRAC sub group 3A) <sup>48</sup>, was approved for use in PT 18 in 2013<sup>49</sup>.

The most sensitive endpoints for deltamethrin are an LD<sub>50</sub> of 0.0015  $\mu$ g/organism for *Apis mellifera* (PPDB) and a LD<sub>50</sub> of 0.0016  $\mu$ g/organism for *Megachile rotundata*, another bee species (Piccolomini et al. 2018). No test results with colleopteran species were available but two endpoints for dipteran species (Sukontason et al. 2005), beeing by a factor of appr. 30 times (blow fly) and 70 times (house fly) less sensible than HB.

Species tested	Order (family)	Test duration (hours)	LD <sub>50</sub> in µg/organis m	Weight normalised LD <sub>50</sub> in µg/g organism	Reference
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	n.a 24	<b>0.0015</b> – 112.2	0.0125 - 935	PPDB; Thompson 2001; del Sarto et al. 2014
<i>Bombus</i> <i>terrestris</i> ssp. audax (buff-tailed bumblebee)	Hymenoptera (Apidae)	72	0.79	n.a.	Reid et al. 2020
<i>Bombus terrestris</i> (buff-tailed bumblebee)	Hymenoptera (Apidae)	n.a.	>0.2	>0.9	PPDB
<i>Megachile</i> <i>rotundata</i> (alfalfa leafcutting bee)	Hymenoptera (Apidae)	n.a 24	<b>0.0016</b> - 0.556	<b>0.0545</b> - 18.91	PPDB; Tassei et al. 1988; Piccolomini et al. 2018
<i>Osmia bicornis</i> (red mason bee)	Hymenoptera (Apidae)	n.a.	0.057	0.61	PPDB
<i>Melipona quadrifasciat a</i> (stingless bee)	Hymenoptera (Apidae)	24	129.2	15,950.62	del Sarto et al. 2014
<i>Musca domestica</i> (house fly)	Diptera (Muscidae)	24	0.1058	n.a.	Sukontason et al. 2005
<i>Chrysomya megacephala</i> (blow fly)	Diptera (Calliphoridae)	24	0.0461	n.a.	Sukontason et al. 2005

**Table 21:** Comparison of acute toxicity endpoints for different species topically exposed to deltamethrin.

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Bombus terrestris ssp. audax (buff-tailed bumblebee)	Hymenoptera (Apidae)	72	0.79	n.a.	Reid et al. 2020
<i>Bombus terrestris</i> (buff-tailed bumblebee)	Hymenoptera (Apidae)	n.a.	>0.2	>0.9	PPDB
<i>Megachile rotundata</i> (alfalfa leafcutting bee)	Hymenoptera (Apidae)	n.a. – 24	<b>0.0016</b> - 0.556	<b>0.0545</b> - 18.91	PPDB; Tassei et al. 1988; Piccolomini et al. 2018
<i>Osmia bicornis</i> (red mason bee)	Hymenoptera (Apidae)	n.a.	0.057	0.61	PPDB
<i>Melipona quadrifasciat a</i> (stingless bee)	Hymenoptera (Apidae)	24	129.2	15,950.62	del Sarto et al. 2014
<i>Musca domestica</i> (house fly)	Diptera (Muscidae)	24	0.1058	n.a.	Sukontason et al. 2005
<i>Chrysomya megacephala</i> (blow fly)	Diptera (Calliphoridae)	24	0.0461	n.a.	Sukontason et al. 2005

#### **Permethrin**

The active substance permethrin, a pyrethroid insecticide (IRAC sub group 3A), was approved for use in PT08 and PT18 in 2016.

For the active substance permethrin, acute contact endpoints after topical application are available for different bee species and species of the orders Lepidoptera and Coleoptera as well as a dipteran species. Based on these endpoints, all tested representatives are sensitive to permethrin, with coleopteran species being the most sensitive ones. When comparing the weight-normalised LD50s, the coleopteran species Adalia bipunctata and the Lepidoptera Heliconius charitonius are the most sensitive, but the endpoints are in the same range as for HB. Lethal doses for ten different coleopteran species are presented, ranging from an 24h-LD50 of 0.12  $\mu$ g/organism for Hippodamia convergens to a 24h-LD50 of 0.0016  $\mu$ g/organism for Oulema melanopus, the latter one by a factor of 75 more sensitive than Hippodamia convergens.

**Table 22:** Comparison of acute toxicity endpoints for different species topically exposed to permethrin.

Species tested	Order (family)	Test duration (hours)	LD <sub>50</sub> in µg/organism	Weight normalised LD <sub>50</sub> in µg/g organism	Reference
<i>Vanessa cardui</i> (painted lady)	Lepidoptera (Nymphalidae)	24	0.24 <sup>49</sup> (1.76 <sup>50</sup> )	<u>1.13</u> <sup>49</sup> (8.26 <sup>50</sup> )	Hoang et al. (2011)
<i>Bombus terricola</i> (yellow-banded bumble bee)	Hymenoptera (Apidae)	48	0.217	1.42	Helson et al. (1994)
<i>Bombus terrestris</i> (buff-tailed bumblebee)	Hymenoptera (Apidae)	n.a.	>0.22	>0.99	PPDB
Junomia coenia (common buckeye) <sup>52</sup>	Lepidoptera (Nymphalidae)	24	0.12 <sup>49</sup> (0.72 <sup>50</sup> )	<u>1.13</u> <sup>49</sup> (6.79 <sup>50</sup> )	Hoang et al. (2011)
Trigona spinipes (spiny-legged stingless bee) <sup>54</sup>	Hymenoptera (Apidae)	24	0.0724	5.17	Macieira and Hebling- Beraldo (1989)
Eumaeus atala (Atala) <sup>52</sup>	Lepidoptera (Lycaenidae)	24	0.20 <sup>49</sup> ( <u>0.07</u> <sup>50</sup> )	1.71 <sup>49</sup> ( <u>0.6</u> <sup>50</sup> )	Hoang et al. (2011)
<i>Eumaeus atala</i> (Atala) <sup>52</sup>	Lepidoptera (Lycaenidae)	24	0.00042	0.0036	Salvato (2001)
<i>Melipona beecheii</i> (stingless bee) <sup>51</sup>	Hymenoptera (Apidae)	24	0.066	1.16	Valdovinos- Núñez et al. (2009)
<i>Anartia jatrophae</i> (white peacock) <sup>52</sup>	Lepidoptera (Nymphalidae)	24	0.06 <sup>49</sup> (0.26 <sup>50</sup> )	0.625 <sup>49</sup> (2.71 <sup>50</sup> )	Hoang et al. (2011)
Megachile rotundata (alfalfa leafcutting bee)	Hymenoptera (Megachilidae)	24	0.057	1.94	Piccolomini et al. (2018)
Coccinella transversoguttata	Coleoptera (Coccinellidae)	24	0.048	1.4	Coats et al. (1979)
<i>Osmia lignaria</i> (orchard mason bee) <sup>52</sup>	Hymenoptera (Megachilidae)	96	0.031	0.279	Peterson et al. (2021)

<sup>&</sup>lt;sup>49</sup> topical application on thorax

 <sup>&</sup>lt;sup>50</sup> topical application on forewings
 <sup>51</sup> not native to Europe, eusocial
 <sup>52</sup> native to North America

<i>Heliconius charitonius</i> (zebra longwing) <sup>52</sup>	Lepidoptera (Nymphalidae)	24	0.03 <sup>49</sup>	0.23 <sup>50</sup>	Hoang et al. (2011)
<i>Hippodamia glacialis</i> (glacial lady beetle) <sup>52</sup>	Coleoptera (Coccinellidae)	24	0.0264	1.2	Coats et al. (1979)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	48	0.021	0.173	Helson et al. (1994)
<i>Apis mellifera</i> (honey bee)	Hymenoptera (Apidae)	48	0.0235	0.196	AR Permethrin PT 18 (ECHA, 2014); DEFRA Report (2008)
<i>Fannia canicularis</i> (lesser house fly)	Diptera (Fanniidae)	n.a.	0.022	n.a. <sup>53</sup>	Meyer et al. (1990)
<i>Trigona nigra</i> (fruit-scarring bee) <sup>54</sup>	Hymenoptera (Apidae)	24	0.021	2.12	Valdovinos- Núñez et al. (2009)
Megachile rotundata (alfalfa leaf-cutting bee)	Hymenoptera (Apidae)	48	0.0176	0.616	Helson et al. (1994); DEFRA Report (2008)
<i>Megachile rotundata</i> (alfalfa leaf-cutting bee)	Hymenoptera (Apidae)	n.a.	0.0157	0.535	PPDB
<i>Hippodamia convergens</i> (convergent lady beetle) <sup>52</sup>	Coleoptera (Coccinellidae)	24	0.012	0.8	Coats et al. (1979)
<i>Nannotrigona perilampoides</i> (stingless bee) <sup>55</sup>	Hymenoptera (Apidae)	24	0.01	1.37	Valdovinos- Núñez et al. (2009)
<i>Hippodamia</i> <i>tredecimpunctata</i> <i>tibialis</i> (thirteenspotted lady beetle)	Coleoptera (Coccinellidae)	24	0.01	0.87	Coats et al. (1979)
<i>Andrena erythronii</i> (trout lily miner bee) <sup>56</sup>	Hymenoptera (Andrenidae)	48	0.0084	0.152	Helson et al. (1994

<sup>53</sup> Information not available
 <sup>54</sup> not native to Europe, eusocial
 <sup>55</sup> neotropical distribution

<sup>56</sup> native to Eastern North America

<i>Coccinella trifasciata</i> (three-banded lady beetle)	Coleoptera (Coccinellidae)	24	0.0048	0.37	Coats et al. (1979)
Adalia bipunctata (two- spotted ladybird beetle)	Coleoptera (Coccinellidae)	24	0.0011	0.10	Coats et al. (1979)
<i>Hippodamia parenthesis</i> (parenthesis lady beetle) <sup>52</sup>	Coleoptera (Coccinellidae)	24	0.0033	0.40	Coats et al. (1979)
<i>Coleomegilla maculata</i> spp. lengi (spotted lady beetle) <sup>52</sup>	Coleoptera (Coccinellidae)	24	0.0021	0.14	Coats et al. (1979)
<i>Diabrotica longicornis</i> (northern corn rootworm)	Coleoptera (Chrysomelida e)	24	0.0017	0.24	Coats et al. (1979)
<i>Oulema melanopus</i> (cereal leaf beetle)	Coleoptera (Chrysomelida e)	24	0.0016	0.22	Coats et al. (1979)
<i>Heliconius charitonius</i> (zebra longwing) <sup>52</sup>	Lepidoptera (Nymphalidae)	24	0.000053	0.0004	Salvato et al. (2001)
<i>Musca domestica</i> (house fly)	Diptera	24	0.0049	1	Sukontaso n et al. (2005)
<i>Chrysomya megacephala</i> (blow fly)	Diptera	24	0.0028	/	Sukontaso n et al. (2005)

### Data analysis

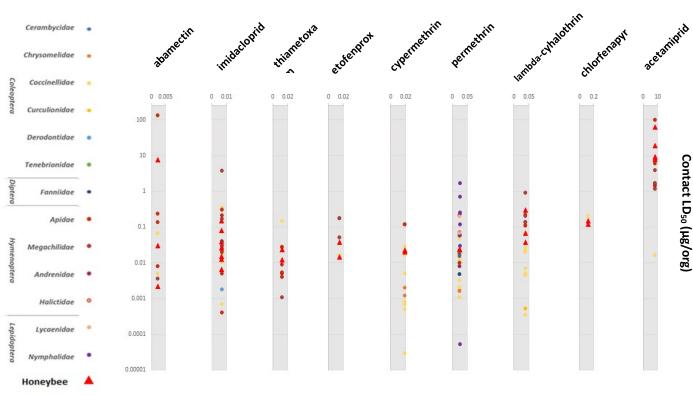
Overall, a data set of 143 datapoints on contact toxicity in adult arthropod pollinators has been gathered across 9 active substances. This data set consists of 83 datapoints for bee and 61 datapoints for NBP. Out of the 83 bee datapoints, 29 correspond to *Apis mellifera* and 13 to BBs species, while the remaining 41 datapoints are spread unevenly across 15 different species of solitary bees. After the Hymenoptera order, the Coleoptera order is the second best represented with a total of 49 datapoints and at least one datapoint for each substance. Data for Lepidoptera and Diptera orders is mostly missing, as combined they account for 11 datapoints overall. In fact, no data on Lepidoptera and Diptera orders has been found for any of the active substances under study except for permethrin.

The distribution of data is uneven across the 9 active substances considered, except for *Apis mellifera*, which was tested for all substances. With 30 and 25 non-honeybee datapoints each, permethrin and imidacloprid are the two most represented substances, while etofenprox and chlorfenapyr account only for 4 and 1 non-honey bee datapoints, respectively. The distribution of non-bee data points is also variable, as permethrin, cypermethrin, lambda-cyhalothrin and imidacloprid account for, in similar amounts, most of the non-bee datapoints, whereas the other

substances account for 0 to 2 datapoints each.

Given the heterogeneous distribution of the dataset across substances and within orders of arthropod pollinators, Species Sensitivity Distribution (SSD) approaches were deemed to be of very limited applicability and therefore unsuitable for a data analysis concerning sensitivity of NBP. However, the gathered data can still provide some key insights on NBP species sensitivity, even though there is a need for caution not to over-generalise conclusions.

NBP species datapoints are found which are up to 3 orders of magnitude lower (e.g., Coleoptera in lambda-cyhalothrin endpoints) than those for HB species, proving that for some substances some NBP species are indeed more sensitive than HB (Figure 10). This figure also illustrates the differences of acute (topical) contact sensitivity based on the endpoints presented in chapter 5. The LD50 (in  $\mu$ g/organism for topical exposure) of different bee and non-bee species are compared to honey bee endpoint(s).



Contact LD<sub>50</sub> (µg/org) for Honey Bees

Figure 10: Distribution of sensitivity endpoints (LD50 in  $\mu$ g/organism) for HB non-bee FVI topically exposed to biocidal active substances.

For abamectin and etofenprox the HB endpoint could cover NBP, as based on the data presented here the  $LD_{50}$  for HB is the most sensitive available endpoint. For other a.s., like permethrin and lambda-cyhalothrin, HB is not the most sensitive species and other bee and non-bee species seems to be more sensitive when topically exposed to the a.s. For these cases, the use of HB sensitivity as a surrogate in the pollinator risk assessment would not be a worst case estimate when considering all non-bee pollinators and appropriate assessment factors could be justified. Given the fact that HBs might be used as a surrogate species for bees, a question that becomes relevant is whether NBP species are found to be more sensitive than bee species. A subset of the data is considered here, which includes bee and coleoptera datapoints for the substances where the existing coverage was deemed acceptable for both groups of species (i.e., imidacloprid, permethrin and lambda-cyhalothrin). Focusing on this subset, it can be asserted that bees and coleoptera show similar sensitivity for imidacloprid, whereas Coleoptera show higher sensitivity for permethrin and lambda-cyhalothrin. Differences of more than one order of magnitude were considered relevant. Noticeably, permethrin and lambda-cyhalothrin belong to a group of substances, the pyrethrins, which have a mode of action different to that of imidacloprid. While the gathered dataset might well be insufficient to draw any conclusions in this regard, it does provide some indication that it will be relevant to incorporate considerations to the mode of action in future research on NBP sensitivity.

No comparison on sensitivity is presented between bees and non-bee orders other than coleoptera, or at a bee/non-bee level of aggregation, due to the already discussed challenges with the distribution of the dataset.

Finally, it is worth noting the range of four orders of magnitude between the three contact  $LD_{50}$  datapoints reported for abamectin endpoints on adult HBs. While the uncertainties associated with effect testing have not been discussed, this example might well illustrate the difficulties involved in the data analysis when different sources of non-standard experiments are considered.

### Conclusion on sensitivity comparison

A non-exhaustive literature review, using e.g. online databases, was done (for details see chapter 5.1). References were chosen if a) the toxicity was given as  $LC_{50}$  or  $LD_{50}$  and b) the test substance was administered topically on test organisms (e.g., to abdomen and/or forewings). The latter was done, as the focus on acute contact endpoints based on topical application allows comparison of  $LD_{50}$  as µg/organism, whereas studies using orally administered test substance not always result in endpoints per organisms, as the amount of ingested test substance per organisms is not recorded for most studies.

The data base presented above is scarce for NBP, especially for Diptera and non-bee Hymenoptera. Species that are vulnerable based on their ecological traits are not always the species available for toxicity tests. Relevant publications of toxicity endpoints are rare or could not be found in the scope of this research or did not fit the criteria as stated above. Relevant endpoints from open literature, data bases or studies used for regulatory purposes were collected and summarized in the attached Annex, in section 4.2 and tables Table 14 to Table 21 above. Although all presented LD<sub>50</sub>s were derived for acute contact exposure by topical application of test substances, some parameters differ, mainly test duration and type of test substance (active substance or formulation).

For Coleoptera, a comparison of acute contact toxicity endpoints (as  $\mu$ g/organism) was done for different active substances (Table 14 to 20). Based on the available data, some coleopteran species seem to be more sensitive than *Apis mellifera* for certain active substances (e.g., imidacloprid, acetamiprid). The same could be assumed for other groups of NBP, as the LD<sub>50</sub>s for different Lepidoptera species exposed to permethrin (Table 21) point to the same direction. For Diptera, only one LD<sub>50</sub> for the lesser house fly (*Fannia canicularis*) exposed to permethrin is shown here, published in Meyer et al. (1990).

Although the limited data set and the lack of statistical analysis on toxicity endpoints, the impression is that some non-bee species seem to be more sensitive to certain active substances when topically exposed. For permethrin, an active substance approved for use in PT08 and PT18, LD<sub>50</sub>s are available for 23 different species from four orders (Hymenoptera, Coleoptera, Diptera and Lepidoptera). This allows some preliminary conclusions, but toxicity data are not available for most of the test organisms exposed to other biocidal active substances.

Both, the lethal doses in  $\mu$ g/organism and the weight-normalised LD<sub>50</sub> ( $\mu$ g/g body weight) are given, as individual body weight and size is known to have an influence on sensitivity of organisms.

Based on the available data presented above, the sensitivity of NBP can be comparable or even higher to the sensitivity of BP for some substances. It remains to clarify which life stage of NBP could be more sensitive, and therefore which are the most relevant route of exposure. For example, some publication found in open literature, suggest that larvae could be even more sensitive than adults (e.g., for beetles), so more information should be gained for this topic.

## 4.3 Data gaps and recommendations for future research

NBP are a highly diverse group of organisms with different ecological traits. As already described in section 4.2, it is still not possible to conclude on sensitivity differences between bee and nonbee species, as information is scarce for all relevant families/species. In this context, it would be highly valuable to have more laboratory studies for acute contact/oral toxicity that compare sensitivity between NBP and HB along similar parameters. Moreover, further studies are needed to find out which is the most relevant route of exposure of NBP and at which life stage they are most exposed to chemicals in environmental conditions to be able to make reliable comparisons and extract the necessary conclusions to develop risk assessment methodologies that cover these organisms.

Nevertheless, based on the data collected for dipteran, lepidopteran, and coleopteran, species and presented in chapter 5.2, it seems that some non-bee species are highly sensitive when exposed to biocidal active substances and may not be covered by a risk assessment targeted on bees only. As this could be assumed also for other NBP groups (e.g., non-bee Hymenoptera), NBP should not be ignored when aiming for the protection of pollinators.

In general, a conclusive evaluation of species sensitivity is needed before we can select suitable test species for NBPs. However, some matters need to be highlighted that are important to consider when in the future when information is available and relevant test species can be selected. Romeis et al. (2013) suggest that "surrogate" species should be selected that most closely meet following the three criteria:

- (i) **Potential sensitivity**: species should be the most likely to be sensitive to the active compound based on the known spectrum of activity of the active ingredient, its mode of action, and the phylogenetic relatedness of the test and target species.
- (ii) **Relevance**: species should be representative of valued taxa or functional groups that are most likely to be exposed to the arthropod-active compound in the field;
- (iii) **Availability and reliability**: suitable life-stages of the test species must be obtainable in sufficient quantity and quality, and validated test protocols must be available that allow consistent detection of adverse effects on ecologically relevant parameters.

In addition, the following criteria should be considered when deciding on a suitable test species as it should:

- be representative of a range of ecosystems
- be easy to maintain and raise in the laboratory (avoidance of wild captures if rearing not possible)
- medium in size to ensure easy handling and less space required for experimental set ups
- be docile enough to develop standardized testing in laboratory, semi-field and field conditions, or there should be a possibility to conduct computer modelling on the species
- have well described biology (feeding habits, reproduction, basic behaviour) as this is critical to be able to interpret results
- learn to respond to a stimulus in the laboratory, and the response is meaningful for risk assessment purposes.

Furthermore, Wallis de Vries et al. (2017) mention that "a widespread European occurrence in field margins, rapid development and rearing experience render three butterfly species suitable as potential candidates for lab and field experiments in future risk assessments: Queen of Spain Fritillary (*Issoria lathonia*), Wall Brown (*Lasiommata megera*) and Swallowtail (*Papilio machaon*)."

In conclusion, all the above criteria should be thoroughly considered when in the future deciding on a suitable test species for assessing the risk to non-bee arthropod pollinators from the use of biocides.

# **5.** Conclusion

In conclusion, with regards to the role of pollination, there is limited information on the extent other species than bees, contribute to pollination. However, when considering the flower visiting frequency, it can be assumed that also NBP significantly contribute to pollination.

The NBP can get exposed to biocidal products during application (e.g., spray mist), by contact to residues (e.g., on plant parts or soil) and/or by uptake of contaminated food (residues in nectar and pollen). Therefore, a possibility of exposure risk can be assumed and NBP should be considered in the risk assessment for biocidal products when developing further guidance.

Although the data base is relatively scarce, NBP show variability in response when exposed to several active substances, and in some cases, they have shown to be as sensitive or even more sensitive than HB.

According to the data collected regarding the sensitivity of these organisms (chapter 5.1), endpoints were only considered relevant if they were given as acute  $LC/LD_{50}$ . A comparison with toxicity data published for bee species was done mainly for Diptera, Lepidoptera, and Coleoptera. The work carried out shows that accurate information on the sensitivity per certain substances is difficult to obtain, as the data base is too small and there are methodological differences in the test designs.

Previous studies (Hoang et al., 2011, 2015; Fernandes et al., 2008; Meyer et al., 1990) compared the LD<sub>50</sub> (µg/organism for topical exposure) of bee and non-bee species for different biocides. In these studies, for some compounds (i.e abamectin and etofenprox) the HB endpoint seemed to cover NBP, showing that the HB LD<sub>50</sub> is the most sensitive available endpoint. However, for other substances (i.e permethrin and lambda-cyhalothrin), HB does not seem to be the most sensitive species and other bee and non-bee species seemed to be more sensitive when topically exposed to the active substance. These findings indicate that although the data base is relatively scarce, there are indications that for some active substances NBP seem to be more sensitive than HBs. In these cases, it seems that the only reference to the sensitivity observed for HB may not ensure, per se, for a comprehensive protection of pollination services in the environment.

Considering the sensitivity of NBP and the ecological traits, would be appropriate for protective strategies for biocidal products in the future. Traditionally, the coverage of risk assessment over multiple species and taxa has been based on testing surrogate species and the application of uncertainty factors to account for inter-specific variability. The results of this review will be taken into account in the further development of guidance for the risk assessment of arthropods pollinators including bees. With this regards it is clear that the risk assessment approach will have to deal with the limited knowledge of the sensitivity variability of NBP, which species and for which substances / mode of action they appear to be more sensitive, in which life stage and what is the relevant exposure pathways. In order to accomplish this, further research would be needed. Furthermore, should testing of other arthropods species than bees be considered, the identification of suitable species would need to respond to multiple criteria, like biological relevance, testing feasibility and standardisation, that would also require further development and analysis.

Due to the high heterogeneity of NBP, further researcher is needed to fill the current data gaps on their ecological traits, their role on pollination and their sensitivity when exposed to biocides. A shift in perspective from a "bee-only focus" to "flower-visiting insect" may be needed to ensure the protection of pollinator biodiversity as well as to better address the whole economic value of pollination. These studies should also consider the services provided by other types of insects, such as flies, wasps, beetles, and butterflies — important pollinators that are currently overlooked.

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## Annex

**Table 23:** Taxonomic overview of relevant FVI in the Phylum Arthropoda, Class Insecta, OrderHymenoptera.

Taxonomic overview of relevant FVI in the Phylum Arthropoda, Class Insecta, Order Hymenoptera				
Common name	Subfamily	Family	Superfamily	Order
Digger wasps		Crabronidae	Apoidea	Hymenoptera
Thread-wasted wasps		Sphecidae	Apoidea	Hymenoptera
cuckoo wasps		Chrysididae	Chrysidoidea	Hymenoptera
Spider wasps		Pompilidae	Vespoidea	Hymenoptera
Scoliid wasps		Scoliidae	Vespoidea	Hymenoptera
Tiphiid wasps		Tiphiidae	Vespoidea	Hymenoptera
Pollen wasps	Masarinae	Vespidae	Vespoidea	Hymenoptera
Paper wasps	Polistinae	Vespidae	Vespoidea	Hymenoptera
Potter wasps	Eumeninae	Vespidae	Vespoidea	Hymenoptera
Paper wasps	Vespinae	Vespidae	Vespoidea	Hymenoptera
Stem sawflies		Cephidae	Cephoidea	Hymenoptera
Serrate-horned sawflies		Megalodontesidae	Pamphilioidea	Hymenoptera
Common sawflies		Tenthredinidae	Tenthredinoidea	Hymenoptera
Argid sawflies		Argidae	Tenthredinoidea	Hymenoptera
Combid sawflies		Cimbicidae	Tenthredinoidea	Hymenoptera

Taxonomic overview of relevant FVI in the Phylum Arthropoda, Class Insecta, Order Diptera				
Common name	Subfamily	Family	Superfamily	Order
Bee flies		Bombyliidae	Asiloidea	Diptera
Hoverflies		Syrphidae	Syrphoidea	Diptera
Housefly/stable fly		Muscidae	Muscoidea	Diptera
Tangle-veined flies		Nemestrinidae	Nemestrinoidea	Diptera
Soldier flies		Stratiomyidae	Stratiomyoidea	Diptera
Horseflies		Tabanidae	Tabanoidea	Diptera
Thick-headed flies		Conopidae	Conopoidea	Diptera
Blow flies		Calliphoridae	Oestroidea	Diptera
Caterpillar flies		Tachinidae	Oestroidea	Diptera

**Table 24:** Taxonomic overview of relevant FVI in the Phylum Arthropoda, Class Insecta, Order Diptera.

**Table 25:** Taxonomic overview of relevant FVI in the Phylum Arthropoda, Class Insecta, OrderLepidoptera.

Taxonomic overview of relevant FVI in the Phylum Arthropoda, Class Insecta, Order Lepidoptera				
Common name	Subfamily	Family	Superfamily	Order
Swallowtail butterflies		Papilionidae	Papilionoidea	Lepidoptera
Pierids		Pieridae	Papilionoidea	Lepidoptera
The Gossamer- wings		Lycaenidae	Papilionoidea	Lepidoptera
Brush footed butterflies		Nymphalidae	Papilionoidea	Lepidoptera
Skippers		Hesperiidae	Hesperioidea	Lepidoptera
Hawk moths		Sphingidae	Bombycoidea	Lepidoptera
Burnet moths		Zygaenidae	Zygaenoidea	Lepidoptera

**Table 26:** Taxonomic overview of relevant FVI in the Phylum Arthropoda, Class Insecta, OrderColeoptera.

Taxonomic overview of relevant FVI in the Phylum Arthropoda, Class Insecta, Order Coleoptera				
Common name	Subfamily	Family	Superfamily	Order
Scarab beetles		Scarabaeidae	Scarabaeoidea	Coleoptera
Ladybird beetles		Coccinellidae	Coccinelloidea	Coleoptera
Sap beetles		Nitidulidae	Cucujoidea	Coleoptera
Shining flower beetles		Phalacridae	Cucujoidea	Coleoptera
Pleasing fungus beetle		Erotylidae	Cucujoidea	Coleoptera
Snout beetles/true weevils		Curculionidae	Curculionoidea	Coleoptera
Long-horned beetles		Cerambycidae	Chrysomeloidea	Coleoptera
Leaf beetles		Chrysomelidae	Chrysomeloidea	Coleoptera
Soft-winged flower beetles		Melyridae	Cleroidea	Coleoptera
Checkered beetles		Cleridae	Cleroidea	Coleoptera
Tumbling flower beetles		Mordellidae	Tenebrionoidea	Coleoptera
Blister beetles		Meloidae	Tenebrionoidea	Coleoptera
False blister beetles		Oedemeridae	Tenebrionoidea	Coleoptera
Jewel beetles		Buprestidae	Buprestoidea	Coleoptera
Soldier beetles		Cantharidae	Elateroidea	Coleoptera
Click beetles		Elateridae	Elateroidea	Coleoptera
Water scavenger beetles		Hydrophilidae	Hydrophiloidea	Coleoptera
Rove beetles		Staphylinidae	Staphylinoidea	Coleoptera

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