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## **Preliminary considerations and planned methods for the revision of Tier 1 risk assessment schemes of EFSA's 2013 guidance on the risk assessment of plant protection products on bees (*Apis mellifera*, *Bombus* spp. and solitary bees)**

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### **DRAFT for the consultation of MSs and Stakeholder Group 20 March 2020**

#### **Abstract**

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## Summary

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## Table of contents

Abstract.....	1
Summary.....	3
PART 1 6	
1. Introduction.....	6
1.1. Background and Terms of Reference as provided by the requestor .....	6
1.2. Overview of EFSA's approach to revising 2013 guidance.....	6
1.3. Scope and structure of this document .....	8
PART 2 9	
2. Outline of the risk assessment process in the revised guidance.....	9
2.1. Target groups.....	9
2.1.1. General information on bee life history .....	9
2.1.2. European Honey bees ( <i>A. mellifera</i> ) .....	9
2.1.3. Bumblebees ( <i>Bombus</i> spp.) .....	9
2.1.4. Solitary bees (multiple genera) .....	10
2.2. Tiered risk assessment scheme .....	10
2.3. Characteristics of the risk assessment schemes.....	11
PART 3 12	
3. Problem formulation for Tier 1 risk assessment schemes .....	12
3.1. Overall risk assessment conceptual model .....	12
3.2. Prioritisation of risk assessment parameters or topics (issues) and related methodological approach.....	12
3.3. Exposure assessment .....	13
3.3.1. Exposure routes and scenarios.....	13
3.3.2. Dietary exposure via pollen and nectar (oral).....	14
3.3.2.1. Dietary model .....	14
3.3.2.2. Prioritisation of the parameters .....	18
3.3.3. Exposure via water consumption (oral).....	19
3.3.4. Contact exposure .....	19
3.4. Crop attractiveness .....	20
3.5. Further considerations.....	20
PART 4 22	
4. Proposed approach for the review of parameters or topics of priority 2 and 3 .....	22
4.1. Exposure assessment .....	22
4.1.1. Succeeding crop scenario .....	22
4.1.1.1. Background of the issue .....	22
4.1.1.2. Proposed approach .....	22
4.1.2. Weeds in field scenario.....	23
4.1.2.1. Background and problem formulation .....	23
4.1.2.2. Methods for data collection.....	24
4.1.2.3. Dataset 1: monitoring study in maize and potato .....	25
4.1.2.4. Dataset 2: weed recordings from control plots of herbicide efficacy trials .....	26
4.1.2.5. The 'significant fraction' of surface area covered by weeds .....	32
4.1.2.6. Preliminary, overall conclusions.....	33
4.1.3. Pre-flowering factor.....	35
4.1.3.1. Background of the issue .....	35
4.1.3.2. Proposed approach .....	37
4.1.4. Protein content of pollen .....	37
4.1.4.1. Background of the issue .....	37
4.1.4.2. Review process that was followed .....	37
4.1.4.3. Preliminary conclusions .....	38
4.1.5. Landscape dilution factor.....	38
4.1.5.1. Background of the issue .....	38
4.1.5.2. Proposed approach .....	39
4.1.6. Exposure via water consumption.....	40

4.1.6.1. Background of the issue .....	40
4.1.6.2. Proposed approach .....	40
4.2. Hazard assessment .....	50
4.2.1. Inter-species toxicity endpoint difference and related assessment factors.....	50
4.2.1.1. Background of the issue .....	50
4.2.1.2. Proposed approach .....	51
4.2.2. Extrapolation factor beyond the tested rates for less toxic substances.....	56
4.2.2.1. Background of the issue .....	56
4.2.2.2. Proposed approach .....	57
4.2.2.3. Slope data collection .....	59
4.2.3. Suitability of the OECD 239 Guidance Document and the expected pesticide residue levels of the processed food of honey bees.....	61
4.2.3.1. Background of the issue .....	61
4.2.3.2. Review process that was followed .....	61
4.2.3.3. Preliminary conclusions .....	65
PART 5	66
5. Planned methods for assessing high priority RA parameters or topics.....	66
5.1. Methods to assess crop attractiveness .....	66
5.1.1. Definitions .....	66
5.1.2. Problem formulation and assessment model .....	66
5.1.3. Crop list.....	68
5.2. Methods to estimate food consumption: systematic review .....	69
5.2.1. Question formulation.....	69
5.2.2. Eligibility criteria for study selection.....	70
5.2.3. Search strategies .....	74
5.2.4. Study selection process .....	75
5.2.5. Evidence appraisal .....	76
5.2.5.1. Summary of the internal validity and external validity of each individual study, by endpoint...	82
5.2.6. Data extraction from the included studies and evidence synthesis.....	83
5.3. Methods to estimate crop-specific sugar content in nectar: systematic review.....	84
5.3.1. Question formulation.....	84
5.3.2. Eligibility criteria for study selection.....	84
5.3.3. Search strategy.....	85
5.3.4. Study selection process .....	86
5.3.5. Evidence appraisal .....	86
5.3.5.1. Presentation of the results of evidence appraisal, by endpoint .....	90
5.3.6. Data extraction from the included studies and evidence synthesis.....	90
5.4. Residue levels in pollen/nectar and TWA .....	91
5.4.1. Background .....	91
5.4.2. Aim of the review.....	92
5.4.3. Question formulation.....	92
5.4.4. Eligibility criteria and evidence appraisal .....	92
5.4.5. Search strategy, study selection and data extraction .....	92
5.4.6. Analysis of the amended database .....	93
5.5. Methods to update the exposure related parameters: $f_{dep}$ and $E_f$ .....	93
References.....	95
Glossary [and/or] Abbreviations .....	100
Appendix A – Sensitivity analysis.....	101
Appendix B – Method for the EKE on crop attractiveness (active pollen collection) .....	108
Appendix C – Search strings on food consumption .....	114
Appendix D – Search strings on sugar content .....	122

## PART 1

### 1. Introduction

#### 1.1. Background and Terms of Reference as provided by the requestor

In 2013, EFSA issued a guidance document on the risk assessment (RA) of plant protection products on bees (*Apis mellifera*, *Bombus spp.* and solitary bees) (EFSA, 2013), which so far has not been fully implemented in the regulatory framework owing to some lack of consensus and request for revision by the Member States.

Based on the above, in March 2019 the European Commission mandated EFSA to revise the guidance (SANTE/E4/SH/gb(2019)1623216). According to the mandate, the review should, in particular:

1. *take account of the feedback from Member States and stakeholders on the EFSA (2013) guidance document (Term of Reference (ToR) 1);*
2. *provide a review and summary of the evidence as regards bee background mortality, in particular considering realistic bee keeping management for *Apis mellifera* and natural background mortality. EFSA is requested to provide this summary in a separate document from the guidance document (ToR2);*
3. *review the list of bee-attractive crops in particular considering presence of bees, guttation and agricultural practices (harvesting time before or after flowering). This reviewed list shall also mention at which growing phases (e.g. BBCH codes) a crop is considered bee-attractive (ToR3);*
4. *review the current risk assessment methodologies in light of recent scientific research and developments e.g. exposure estimation, relevance of the exposure scenarios (e.g. weed scenario) and relevance of some risk assessment schemes. Available relevant guidance developed by Member States should be considered (e.g. draft Guidance Document on seed treatments and/or its follow up work) (ToR4);*
5. *review the requirements for higher tier testing, in particular by reconsidering the magnitude of detectable effects vs the statistical power and validated population modelling in light of realistic agro-environmental conditions (ToR5);*
6. *take into account planned and on-going discussions initiated by the Commission on defining specific environmental protection goals and review the risk assessment guidance based on the specific protection goals agreed during this process (ToR6).*

#### 1.2. Overview of EFSA's approach to revising 2013 guidance

The scientific approach to revising the EFSA, 2013 is tailored to the various ToRs of the mandate. Procedural aspects were explained in the outline published in July 2019 and updated in February 2020<sup>1</sup>.

1. ToR 1 (feedback from Member States and stakeholders on the EFSA, 2013)

Over the summer of 2019, a written procedure took place where MSs and stakeholders of the ad hoc group were invited to comment the EFSA, 2013. This resulted in a comprehensive list of issues/comments/ideas that was used as a basis to plan the guidance revision process and prioritise topics, as explained in Section 3.2 of this document.

<sup>1</sup> [https://www.efsa.europa.eu/sites/default/files/event/Bee\\_Guidance\\_review.pdf](https://www.efsa.europa.eu/sites/default/files/event/Bee_Guidance_review.pdf)

## 2. ToR 2 (bee background mortality)

In 2019, EFSA prepared a separate protocol illustrating the plan for collecting, appraising and synthesising evidence on bee background mortality. In October 2019, Member States and the stakeholders of the ad hoc group were consulted to provide feedback and input to that draft plan. The protocol was revised accordingly and implemented immediately after. The results will be included in a technical report on bee background mortality which is in preparation and will be published.

## 3. ToR 3 and ToR 4 (review of list of bee-attractive crops and of current risk assessment methodologies):

The ToR3 and ToR4 are specifically considered in this document. In particular the EFSA WG describe here the problem formulation and the proposed approaches for revising the crop attractiveness to pollen and nectar and the risk assessment methodologies, focusing on the Tier 1 risk assessment schemes.

As for exposure estimation of dust generated from solid formulations of plant protection products, any update will depend on revision and finalisation of the available Draft Guidance Document for the Authorisation of Plant Protection Products for Seed Treatment (SANCO/10553/2012, July 2018\_rev 16), still ongoing.

## 4. ToR 5 (review of the requirements for higher tier testing)

This ToR5 will be the subject of a separate sub-project that will be carried out within this mandate at a later stage of the review process. The work on these aspects will start when agreed Specific Protection Goals (SPGs) are available and frameworks for Tier 1 and Tier 2 risk assessment schemes are advanced.

## 5. ToR 6 (guidance revision based on updated SPGs)

Specific Protection goals (SPGs) for non-target organisms are under definition by the European Commission. EFSA is requested to consider this ongoing activity. The review of the EFSA, 2013 for this aspect will be performed based on that update.

It is noted that, in the EFSA, 2013, SPGs have been defined (and agreed by Risk Managers) following the EFSA scientific opinion (EFSA PPR Panel, 2010) which gives a framework based on the ecosystems service approach. The same approach is considered in the ongoing activity on SPGs of the European Commission. In the EFSA, 2013, the identified ecosystems services that may be impacted following the exposure of bees to pesticides are: pollination, food, genetic resources, cultural service. The various agreed dimensions defining the SPGs able to protect these ecosystems services, reported in the EFSA, 2013, are:

- Service Providing Unit (SPU): honey bees, bumble bees and solitary bees;
- Ecological entities: Colony (honeybees, bumble bee), population (solitary bees)
- Attribute: Colony strength (honey bees, bumble bee), population abundance (solitary bees)
- Magnitude: negligible effect i.e. <7% colony reduction. The colony size reduction of the exposed colonies should be no more than 7% smaller than the control colonies at any time.
- Spatial scale: edge of field
- Temporal scale of protection: at any time
- Degree of certainty: not defined

The above SPGs were implemented in the risk assessment schemes by using a quantitative methodology (see Appendix M of the EFSA, 2013) in order to obtain trigger values for the lower tiers able to be compliant with the SPGs. This methodology is based on a simple

population model (e.g. Khoury model), which was used to link the colony size reduction <7% to forager mortalities; on the real background mortality (see ToR2) and on a linear dose-response relationship. **Once the risk managers have clarified the SPGs, it is considered part of the mandate to review the abovementioned methodology.**

### 1.3. Scope and structure of this document

The aim of this document is to consult at an early stage the Members States via PSN and the Stakeholder Group on the approach for revising Tier 1 schemes for the new EFSA guidance on the risk assessment of plant protection products on bees (*Apis mellifera*, *Bombus* spp. and solitary bees), with a focus on the mandate ToRs 3 ('attractiveness') and 4 ('risk assessment methodologies').

To this end, the document provides an outline of the overall risk assessment process that will be considered in the new guidance (PART 2), and then focusses on the problem formulation for Tier 1 risk assessment schemes, the related exposure scenarios (including crop attractiveness), the risk assessment parameters and the priority assigned to each of them (PART 3), according to the criteria describe in Section 3.2. Based on the latter, a series of preliminary considerations are made for medium- and low-priority aspects (PART 4) and a plan for estimating high-priority parameters or topics is illustrated (PART 5)

Overall, this document was conceived based on the principles and process defined in a project aimed to further improve EFSA's scientific assessment processes (EFSA, 2015) and following the recommendations for protocol development of EFSA's Scientific Committee.<sup>2</sup>

The revision of the parts of the EFSA, 2013 included in this protocol will be finalised accounting for the feedback and input received through the consultation process as appropriate, in order to produce a document that is fit-for-purpose. Deviations from the planned methods may occur during the drafting process and will be documented.

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<sup>2</sup> Endorsed on 20/02/2020 (to be published in April 2020).



## PART 2

### 2. Outline of the risk assessment process in the revised guidance

Like EFSA, 2013, the revised guidance will outline a process by which plant protection products can be evaluated for the risk they may pose to bees.

#### 2.1. Target groups

The target groups for this risk assessment are honey bees (*Apis mellifera*), bumble bees (*Bombus* spp.) and solitary bees. These three groups have differing life histories, so we include a short summary of the major differences between the groups of bees below

##### 2.1.1. General information on bee life history

Eusocial bees, which in Europe are honeybees and bumblebees, produce three types of adult bee, or caste; workers, drones, and queens. The workers caste is the most abundant type of adult and consist of female bees which will usually not reproduce but will forage for food and maintain the nest. Drones and queens are the reproductive castes. Drones are males that leave the colony to reproduce. A queen is a reproductive female that establishes or allows a colony to continue. Solitary bees do not have a sterile worker caste so can just be referred to as males and females.

##### 2.1.2. European Honey bees (*A. mellifera*)

Honey bees are eusocial bees that live in large (1000s-10,000s individuals) perennial colonies with a single egg-laying queen. Whilst non-managed, feral, colonies exist, honeybees in Europe are a managed species and mostly nest in artificial hives. Honeybee nests consists of wax structures made of hexagonal cells, known as combs, that are used to store food and rear young. Honeybees have a highly structured social system. The queen specialises in laying eggs, whilst workers exhibit temporal polyethism, which means tasks are allocated by age; younger workers tend the developing young and maintaining the colony whilst older workers forage for pollen, nectar and water. Honeybee nests contain large reserves of food, which allows the colony to persist throughout the winter and through sustained periods of poor weather.

##### 2.1.3. Bumblebees (*Bombus* spp.)

Bumblebees are eusocial bees that live in small (10s-100s individuals) annual colonies with a single egg-laying queen or parasitic bees that parasite on the eusocial species. Some bumblebee species, primarily *B. terrestris* in Europe, are used as managed pollinators but most species and colonies are wild. Bumblebees also construct their nest structure from wax but lack the regular structure and appearance of honeybee nests. Unlike in honey bees, each queen is responsible for establishing a colony, laying eggs, and foraging for pollen and nectar until the first generations of workers develop. After the first generations of larvae become adults, the queen remains in nest to lay eggs. There is some evidence that bumblebee workers exhibit alloethism, where tasks are allocated by body size, with smaller workers tending the developing young and maintaining the colony whilst larger workers forage for pollen and nectar; however, the distinction between in hive and forager workers is less clear cut than for honeybees. As bumblebee colonies do not persist over winter the

colony stores much less food than honeybees, usually only enough food to allow the colony to persist through short periods of poor weather. If the colony is successful, new queens are produced during the end of the colony cycle. The new queens are the only ones that survive hibernating during the winter and start new colonies the following spring.

#### 2.1.4. Solitary bees (multiple genera)

Solitary bees are a taxonomically diverse group so generalisations may not apply to all species, however, this group is not eusocial. Solitary bees may be univoltine or multivoltine but only a few species overwinter as adults. Only a small number of solitary bee species are managed (*Osmia* spp., [others]) so the vast majority of the (check No., around ~17,000) solitary bee species globally are wild. Solitary bees use a wide variety of nesting substrates (soil, wood, masonry, leaf and other vegetation) and generally provision nests only once, producing a relatively small (10's) number of offspring.

EFSA, 2013 focusses on the colony (honey bees, bumble bees) or the population (solitary bees), the current ecological entity and the attribute to protect according to the framework for defining SPG (see section 1.2).

### 2.2. Tiered risk assessment scheme

The new guidance will also continue to propose a tiered risk assessment scheme starting with simple and cost-effective lower tiers and moving to more complex higher tiers. Each tier will have to ensure that the appropriate level of protection is achieved.

The following tiers were proposed in EFSA, 2013

1. **Screening Tier**, in the EFSA, 2013 this is a simplified version of Tier 1 by e.g. covering all relevant scenarios and considering only one single worst case; The WG will further consider this.
2. **Tier 1**, which combines exposure estimations based on default parameters with laboratory toxicity endpoints. As in EFSA, 2013, Tier 1 will consider different scenarios according to the potential source of the exposure. Tier 1 schemes are the focus of this document;
3. **Tier 2**, which combines exposure estimation based on measured parameters with laboratory toxicity endpoints. This Tier will be relevant for all scenarios that for which a low risk could not be demonstrated at the Tier 1;
4. **Tier 3**, which is based on effect studies conducted in realistic exposure conditions.

Generally, the lower Tiers are intended to sift out plant protection products/uses which pose a low risk and hence prevent unnecessary further testing. Risk assessments conducted according to the lower Tiers will result in Risk Quotients (RQ) separate for acute toxicity, chronic toxicity and toxicity to larvae. RQs will be calculated by dividing the exposure estimation by the toxicity endpoints. The Risk Quotients will be compared to trigger values that reflect the SPGs. RQs lower than the respective triggers will indicate a low risk. Where a RQ is not lower than the trigger, the risk assessment should be continued applying higher Tier step(s) and/or identifying or mitigation measures able to reduce the exposure without generating further evidence.

### 2.3. Characteristics of the risk assessment schemes

The lower tier risk assessments proposed by EFSA, 2013 combined the aspects listed below. During the revision process, it will be considered whether they are suitable for the new risk assessment schemes and if necessary they will be revised.

- Three groups of bees (honey bees, bumble bees, solitary bees);
- Three application methods (Upward/sideward, downward spray, seed treatment, granules);
- Three exposure routes (contact, dietary (oral) via pollen and nectar, oral via water consumption);
- Three types of effects (acute, chronic, larvae) focusing on mortality/emergence failure and some considerations for sublethal effects and accumulative toxicity for honey bees;
- Five scenarios for the dietary exposure and three scenarios for contact exposure and exposure from water consumption.

## PART 3

### 3. Problem formulation for Tier 1 risk assessment schemes

This section illustrates the model that will be applied in the revised guidance for Tier 1 risk assessment schemes, along with the related exposure scenarios (including crop attractiveness), risk assessment parameters and priority assigned to each of them.

#### 3.1. Overall risk assessment conceptual model

As in the previous guidance, the conceptual model for Tier 1 risk assessment schemes will be represented by the 'Risk Quotient = Exposure/Toxicity', i.e. the ratio of the estimated exposure and the plant protection product's toxicity to bees (e.g. the LD<sub>50</sub>).

In this equation the numerator depends on the type of exposure and is detailed in section 3.3, while the denominator is represented by toxicity endpoints derived from laboratory tests such as LD<sub>50</sub>, LDD<sub>50</sub> or NOED (median lethal dose, median dietary lethal dose or no observed effect dose). The current Working Group (WG) proposed some slight amendments regarding this basic equation which are the following:

- it was noted that in some parts of the EFSA, 2013 LC<sub>50</sub> was mentioned as the chronic toxicity endpoint in place of LDD<sub>50</sub> and NOEC was mentioned in place of NOEL. The WG proposes that such equations should refer always to the intake in terms of mass and not to the concentration(s) used in the test
- in place of the term 'NOEL' consistently NOED should be used (i.e. 'dose' instead of 'level')

As a result, the equation proposed by the WG is:

$$\text{Risk Quotient} = \frac{\text{Exposure estimation}}{\text{LD}_{50} \text{ or LDD}_{50} \text{ or NOED}}$$

The Risk Quotient corresponded to the exposure toxicity ratio (ETR) for risks by oral exposure or hazard quotient (HQ) for risks by contact exposure. At a later stage the WG will consider unifying these two terms (e.g. to consider if both could be ETR).

#### 3.2. Prioritisation of risk assessment parameters or topics (issues) and related methodological approach

The risk assessment parameters feeding into the equation illustrated above as well as topics or scenarios that complement the model, including the crop attractiveness, are described in the following sections on problem formulation.

A relative priority was assigned to each of them as either high (1), medium (2) or low (3), based on a combination of the following aspects:<sup>3</sup>

- Impact of the parameter/topic on the model (sensitivity of the model to the parameter and uncertainty and plausible variability of the parameter). For some parameters and topics, their impact on the model was assessed through a sensitivity analysis (Appendix A);

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<sup>3</sup> Based on EFSA, 2010 (*EFSA guidance on systematic review*)

- Sensitivity to interest: stakeholders may have interests related to the parameters of the model (e.g. adherence to pessimistic scenarios out of a precautionary principle or adherence to increased realisms of a certain parameter);
- Intersubjectivity: variability among preliminary expert opinions about the parameter.

Based on the relative priority assigned to the parameters and topics, the WG developed preliminary considerations and proposals on the approaches to review of them.

For the risk assessment parameters or topics deemed of medium or low priority, the WG considered to apply narrative approaches based on their knowledge and expertise. These are illustrated in PART 4.

For risk assessment parameters or topics identified as of high priority, the WG proposed to apply more systematic approach (PART 5). Examples of these approaches are systematic review (SR) for parameters for which it was considered that sufficient data exist and will be available, or formal expert knowledge elicitation processes (EKE) for parameters for which data are limited.

### 3.3. Exposure assessment

#### 3.3.1. Exposure routes and scenarios

The exposure routes that will be reviewed in the new guidance are the ones considered in the EFSA, 2013 namely:

- oral via pollen and nectar consumption (dietary);
- oral via water consumption;
- contact via contamination to spray or dust drift deposition.

The WG considered the possibility to include further exposure routes for the three bee groups i.e. pesticide residues in honey dew, soil, leaves/plant surface, propolis and wax, and it was agreed that these will not be covered by the current review, nevertheless certainly recognised as recommendations for future activities.

In EFSA, 2013, different exposure scenarios depending on the route of exposure, space and time were identified for Tier 1 risk assessment schemes. They are summarised in Table 1.

For the new guidance, the WG deemed to review the relevance of some of those scenarios, i.e. the weeds in the field (section 4.1.2) and succeeding (section 4.1.1) crop scenarios with a priority 2 (medium) or 3 (low), according the criteria given in section 3.2.

**Table 1. Scenarios as considered in the EFSA, 2013 guide document**

Scenario	Contamination route
Dietary exposure (i.e. oral exposure via pollen and nectar consumption)	
Treated crop	Bees collecting pollen and/or nectar from the treated crop and carry back to the colony/nest
Weeds in the field	Bees collecting pollen and/or nectar and carry back to the colony/nest from the weeds unintentionally contaminated or intentionally treated

Scenario	Contamination route
Dietary exposure (i.e. oral exposure via pollen and nectar consumption)	
Field margin	Bees collecting pollen and/or nectar and carry back to the colony/nest from the field margin plants unintentionally contaminated
Adjacent crop	Bees collecting pollen and/or nectar and carry back to the colony/nest from the adjacent crop unintentionally contaminated
Succeeding crop	Bees collecting pollen and/or nectar and carrying it back to the colony/nest from the succeeding crop unintentionally contaminated through the soil
Exposure via water consumption (oral)	
Guttation	Bees collecting guttation water from the treated crop and carry back to the colony
Puddle	Bees collecting water and carry back to the colony from puddle water unintentionally contaminated
Surface water	Bees collecting water and carry back to the colony from surface water unintentionally contaminated
Contact exposure	
Treated crop	Bees visiting the treated crop
Weeds in the field	Bees visiting weeds in the field unintentionally contaminated or intentionally treated
Field margin	Bees visiting plants on field margin unintentionally contaminated

### 3.3.2. Dietary exposure via pollen and nectar (oral)

#### 3.3.2.1. Dietary model

In EFSA, 2013, dietary exposure via pollen and nectar consumption, for all the dietary scenarios, was calculated based on the numerator in the model below (EFSA, 2013):

$$ETR = \frac{AR \times Ef \times SV \times twa}{toxicity\ endpoint}$$

[eq.1]

Where: ETR = exposure toxicity ratio  
 AR = application rate (kg/ha)  
 Ef = exposure factor (-), i.e. proportion of the applied chemical that causes exposure to the relevant matrix  
 SV = shortcut value; residue intake expressed as mass/bee/day or mass/larva/developmental period, such as µg/bee/day or µg/larva/developmental

period for an application rate of 1 kg/ha or 1 mg/seed (for the treated crop scenario for seed treatment)

*twa* = time weighted average (-), i.e. a factor expressing the yearly maximum time weighted average pesticide intake over a relevant period, such as the period that is considered for chronic assessments

With regard to the SV in the 2013 model, this is a multifactorial value calculated as follows:

$$SV = \frac{(CONC_p \times CONS_p) + \left( CONC_n \times \frac{CONS_{sugar}}{sugar\ in\ n} \right)}{1000}$$

[eq.2]

Where: *CONC* = concentration in the matrix, nectar (subscript *n*) or pollen (subscript *p*) after a single application of 1 kg/ha i.e. RUD (mg/kg)  
*CONS* = consumption (mg/bee/day or mg/larva/developmental period in days) of nectar (subscript *n*) or pollen (subscript *p*)  
*sugar in n* is the sugar content in nectar expressed as mass/mass (e.g. kg/kg)

The numerator of [eq.1] represents the pesticide intake of larvae or adult bees. They are the yearly maxima of daily values for the acute risk assessment and the yearly maxima of time weighted average values over a 10-day exposure period for adult bees and over the developmental period for larvae (different time frames for the different group of species) for one spatial unit, i.e. a colony or population at the edge of a treated field. For adults the pesticide intake is the average of all adult bees (e.g. the average of all foragers and average of all nurses for honey bees).

In the EFSA, 2013, the *twa* factors of 0.72 and 0.85 were considered based upon a default pesticide dissipation half-life, DT50, of 10 days and a 10-day time window (chronic exposure to adults) or a 5-day time window (chronic exposure to honey bee larvae), respectively.

For the revised guidance, the WG agreed upon a number of changes to the above model:

- 1) to include the TWA factor into the SV formula
- 2) to account for the possibility that multiple applications (both before and during the flowering if happens) may build up the residues levels in the relevant matrices
- 3) to take into account that the SVs represents residue intake from situations when the pesticide application happens during the flowering and this can lead to unrealistic exposure estimation for pre-flowering applications
- 4) to take into account potential dilution in the residue levels entering the hive arising from the foraging strategy of the bees
- 5) the exposure via soil uptake of the plant will be considered separately (which lead to a different formula for the succeeding crop scenario)

It should be noted that when considering these changes, only the treated crop and the succeeding crop scenarios were in the focus of the working group. After the consultation on this protocol, a careful consideration will be made regarding how these formulas can be adapted to the weed scenario or to the off-field scenarios. As regards to the succeeding crop scenario, the formula to be considered is reported further below.

As a consequence of these proposals, the agreed model to be considered in the reviewed document for the treated crop scenario is the following:

$$ETR = \frac{AR \text{ Ef } ((MAF_{bf} \text{ PFF } SV_{flower} \text{ TWA}_{df}) + (MAF_{df} \text{ TWA}_{df} \text{ SV}_{flower}))}{\text{toxicity endpoint}} \quad [\text{eq.3}]$$

$$SV_{flower} = \frac{(LDF_p \text{ CONC}_p \text{ CONSp}) + \left( LDF_n \text{ CONC}_n \frac{CONS_{sugar}}{\text{sugar in } n} \right)}{1000} \quad [\text{eq.4}]$$

*Where:*  $SV_{flower}$  = shortcut value for flower; residue intake expressed as mass/bee/day or mass/bee larva/developmental period, such as  $\mu\text{g}/\text{bee}/\text{day}$  or  $\mu\text{g}/\text{larva}/\text{developmental period}$  for an application rate of 1 kg/ha or 1 mg/seed (for the treated crop scenario for seed treatment)

$MAF_{df}$  = multi-application factor during flowering (-); expresses the build-up of residue levels in pollen and nectar due to the multiple applications that are performed during the flowering period. When no application is performed during flowering,  $MAF_{df}$  of 0 will be considered.

It is noted that the WG considered that normally only a low number of applications are performed during the flowering period (e.g. 1-4) in which case developing a look-up table for these parameter will be sufficient. For higher number of applications (also depending on the application interval) a time-moving window approach could be necessary ( $MAF_{df} \times TWA_{df}$ ). In those (rare) cases the risk assessments may need to be moved to Tier 2, where these considerations will be required.

$MAF_{bf}$  = multi-application factor before flowering (-); expresses the build-up of residue levels in pollen and nectar due to the multiple applications that are performed before the flowering period. When no application is performed before flowering,  $MAF_{bf}$  of 0 will be considered.

$TWA_{df}$  = factor converting the daily maximum exposure into the average exposure for the specified dissipation rate and time window (-).

It is noted that the WG considered that for Tier 1, a single, common worst-case dissipation value will be agreed for pollen and nectar and that will be considered for the  $TWA_{df}$  calculations. For the Tier 2 risk assessment the dissipation values for pollen and nectar might be demonstrated to be different. In order to account for this potential differences, the above formula for Tier 2 will be adapted accordingly.

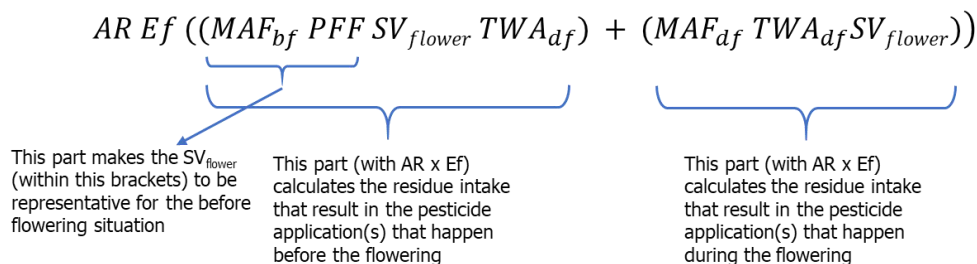
LDF = Landscape dilution factor for pollen (subscript  $p$ ) and nectar (subscript  $n$ ) (-); this factor accounts for potential dilution in the residue levels entering the hive. It is noted that the incorporation and use of this factor is pending on the residue data set to be considered for the SVs. If the residue data set considered in the SVs reflecting the residues entering the hive/nest, then this factor will be considered as redundant. If residues measured directly from the flowers in the field will be considered then this factor is relevant. Further explanations on this factor are included in section 4.1.5.



PFF = pre-flowering factor (-), this factor express that the level of contamination of the nectar and the pollen (i.e. the concentration of the residues in these matrices) resulting from a pesticide application performed during the flowering are different from a pesticide application performed before the flowering. Further explanations on this factor are included in section 4.1.3.

All the other parameters are the same as above [eq.1] or [eq.2].

Similarly to the 2013 version, the numerator of [eq.3] represents the pesticide intake of larvae or adult bees. Some further illustrations/explanations of the numerator are presented below:

$$AR \ Ef \ ((MAF_{bf} \ PFF \ SV_{flower} \ TWA_{df}) + (MAF_{df} \ TWA_{df} \ SV_{flower}))$$


This part makes the  $SV_{flower}$  (within this brackets) to be representative for the before flowering situation

This part (with  $AR \times Ef$ ) calculates the residue intake that result in the pesticide application(s) that happen before the flowering

This part (with  $AR \times Ef$ ) calculates the residue intake that result in the pesticide application(s) that happen during the flowering

As in the EFSA, 2013, most of the parameters above will be scenario/crop type/application type specific and they will be presented in suitable look-up tables. For example the  $Ef$  values, as in the current guidance document, will be by default 1 for the treated crop scenario, but when the formula will be adapted, for example, to the weed scenario, they will reflect the fractions of the pesticide mass that deposits to the weeds and they will be application type specific.

As regards to the succeeding crop scenario, the agreed model to be considered in the reviewed document is the following:

$$ETR = \frac{PEC_{pw} \ SF \ SV_{soil}}{\text{toxicity endpoint}}$$

[eq.5]

$$SV_{soil} = \frac{(LDF_p \ CONS_p) + (LDF_n \ \frac{CONS_{sugar}}{sugar \ in \ n})}{1000}$$

[eq.6]

Where:  $PEC_{pw}$  = concentration in the soil pore water at around of the flowering time of the succeeding crop/permanent crop in the next year (mg/L i.e. ~ mg/kg)  
 This concentration/these concentrations can be obtained by using a specific software package and will be routinely available in pesticide dossiers. For further details please consult section 4.1.1.

SF = safety factor, accounts for the uncertainty of the link between the pore water concentration and the expected concentrations in pollen and nectar

$SV_{\text{soil}}$  = shortcut value for soil; food consumption expressed as mass/bee/day or mass/larva/developmental period, such as mg/bee/day or mg/larva/developmental period

All the other parameters are the same as above [eq.2] or [eq.4].

### 3.3.2.2. Prioritisation of the parameters

The risk assessment parameters that feed into this model, their relative priority (defined based on the considerations made in section 3.2) and the outline of the method that will be applied for estimating them are summarised in Table 2. The Table also points to the relevant sections where these parameters are extensively defined and addressed.

This model will be applicable to the three groups of bees at all life stages and all scenarios. However, the values of the parameters will be case-specific (e.g.  $TWA_{df}$  for all acute assessments will be 1, but may be < 1 for some chronic assessments; LDF for solitary bees will be 1, but may be < 1 for honey bee pollen for the treated crop scenario).

**Table 2. Dietary exposure via pollen and nectar: risk assessment parameters, relative priority and outline of the method for estimating them**

Risk assessment parameter <sup>a</sup>	Relative priority High (1); Medium (2); Low (3)	Approach (PART in this document) <sup>b</sup>
Pre-flowering factor	2	Narrative review/Expert judgment (4)
Food consumption (i.e. pollen and nectar consumption by the bee species living in the EU)	1	Systematic review (5)
Sugar content of the nectar of the different crops grown in the EU	1	Systematic review (5)
Protein content of pollen	3	Narrative review/Expert judgment (4)
Residue levels in pollen/nectar (range of residues on the plants and range of residues entering the hive/nest)	1	Update of existing database with studies that already collected and reported in an external report of EFSA (dossier data) (5)
Time Weighted Average during flowering ( $TWA_{df}$ ) (expressing the range of half-lives of pesticides in pollen and nectar)	1	Same as for residues (5)
Landscape dilution factor	2	Narrative review/Expert judgment (4)
Ef factor	1	Update of existing values with an already reported table of EFSA (5)

<sup>a</sup> Extensive definitions given in the relevant sections below.

<sup>b</sup> PART 4: 'Preliminary considerations on risk assessment parameters or topics of medium and low priority'; PART 5: 'Planned methods for assessing high-priority risk assessment parameters or topics'.

### 3.3.3. Exposure via water consumption (oral)

In EFSA, 2013, exposure via water consumption is calculated considering the numerator of the formula below:

$$ETR = \frac{W \text{ PEC}}{\text{toxicity endpoint}}$$

Where: ETR = exposure toxicity ratio

W = water consumption expressed as volume/bee/day or volume/larva/developmental period, such as  $\mu\text{L}/\text{bee}/\text{day}$  or  $\mu\text{L}/\text{larva}/\text{developmental period}$

PEC = predicted environmental concentration, the concentration of the chemical in water expressed in  $\mu\text{g}/\mu\text{L}$

In EFSA, 2013 three scenarios for water resources were included: guttation water, puddle water and surface water. The WG deemed to consider this topic as medium priority. A preliminary consideration on the relevance of the exposure via water consumption is reported in section 4.1.6 along with proposed approaches to address and review this issue. Changes on the above model will depend on the outcome of this review.

### 3.3.4. Contact exposure

In EFSA, 2013 document, the contact exposure was calculated based on the numerator of the formula below:

$$HQ = \frac{AR \times f_{dep}}{\text{toxicity endpoint}}$$

Where: HQ is the hazard quotient

AR is the application rate (g/ha)

$f_{dep}$  is the deposition factor (-), relevant for the weed scenario and the field margin scenario

No particular concerns were raised by MSs, the stakeholder Group and the WG as well regarding the risk assessment scheme for the contact route of exposure. The only exception was a consideration by the WG to introduce a multi-application factor into the model. This was however finally concluded as not reasonable considering the lifespan and the foraging strategy of the different forager bees (it was considered that over-spaying the same individual, especially in notable number of them, more than ones with the same pesticide in the same field would lead to a rather extreme scenario). Therefore, currently no particular amendments were planned.

The following issues are however noted:

- The crop interception table that was considered for the  $f_{dep}$  parameter was updated after 2013. Therefore, the respective figure will also be updated according to this change (see section 5.5)
- In the context of the planned activity for the exposure assessment on dust drift, the WG will or will not reconsider some of the factors currently used in the risk assessment schemes for solid formulations. It is noted that, the risk assessment schemes for contact exposure, including the exposure via dust, will be part of the revised guidance, while for the exposure characterisation reference will be made to

Draft Guidance Document for the Authorisation of Plant Protection Products for Seed Treatment (SANCO/10553/2012, July 2018\_rev 16).

### 3.4. Crop attractiveness

As for **crop attractiveness** (mandate ToR3), owing to its impact on the model (if a crop is not attractive, then exposure is negligible), the WG deemed to consider this aspect as **priority 1** (high) to estimate through a formal expert knowledge elicitation process. Details on the definition of crop attractiveness as agreed by the WG and the planned methods for assessing via EKE it are given in section 5.1

### 3.5. Further considerations

It should be noted that this protocol does not comprehensively outlines all the changes and amendments what will, potentially, made regarding the Tier 1 risk assessment. There are additional issues what the WG may consider at a later stage. For example such questions like: what should be the list of the likely rotational crops (i.e. make the succeeding crop scenario more realistic), what should be the list of the likely crops that can be in flowering stage at the vicinity of the treated field when the pesticide application happens (i.e. make the adjacent crop scenario more realistic), how often the off-field habitat is dominated by two dimensional structure rather than consists of shrubs and trees; should not the two off-field scenarios merged into one scenario.

Both the consultation on the EFSA, 2013 and the regulatory experience by using this guidance document resulted in some issues which might be considered by the WG at a later stage (e.g. relevance of the weed scenario for seed treatment). The risk assessment schemes for the metabolites and the recommendations for sublethal effects will also be reviewed (those considerations are not part of this protocol).

However, some of those issues, not necessary belonging to the exposure or the exposure model as described above, are already under consideration and some descriptions are available in this protocol.

The relevance of the weed scenario had already been considered and the outcome of that work (as it is at the moment of the publication of this protocol) is outlined in 4.1.2.

Also, the residue levels in the processed food in comparison to the residue levels of pollen and nectar had been investigated. Although this was considered rather in the context of the suitability of a particular test guideline, this is useful also to understand better some mechanism of the exposure of honey bees. That consideration is outlined in 4.2.3.

As regards to the hazard characterisation, some works are also planned. These are outlined in sections 4.2.1 and 4.2.2.

In addition, some pragmatism will be considered by the WG to make the risk assessment schemes and the guidance document more `user friendly`. For example, it will be considered if the terminology used in the different exposure routes could be harmonized (e.g. HQ is used for contact, but ETR for oral route of exposure;  $f_{dep}$  is used in the contact scheme, but for similar parameters Ef is used for the dietary route of exposure). It is also noted that the calculated ETR values of the dietary schemes are usually resulting in some very low numbers (e.g.  $\ll 0.0$ ) making the interpretation of the outcome of the risk assessment difficult. By using a multiplication factors of 100 or 1000 for both the exposure and the respective trigger value, would not change the ratio of the ETR and the trigger value (i.e. no mathematical alteration in the meaning of the risk assessment), but would bring the

ETR values closer or even above 0 (i.e. into a more comfortable range). This could be done e.g. to swap the unit of the application rate from kg/ha to g/ha (which would mean again a harmonization with the contact scheme).

## PART 4

### 4. Proposed approach for the review of parameters or topics of priority 2 and 3

This section illustrates the proposed approach to addressing topics or issues deemed of priority 2 (medium) and 3 (low), as explained in section 3.2. For the topics outlined in sections 4.1.2, 4.1.4 and 4.2.3, some preliminary conclusions are also made.

#### 4.1. Exposure assessment

##### 4.1.1. Succeeding crop scenario

###### 4.1.1.1. Background of the issue

For the succeeding crop scenario it is considered that the residues of the substance that are already present in the soil are taken up by the roots of the permanent crops next year or in succeeding annual crops and translocated via the vascular system and the tissues of plants to the nectar and pollen. In the current guidance it is considered that if the succeeding crops are not defined (which is the case in the GAP tables for EU evaluation), it is assumed that the crops are attractive for both the pollen and nectar.

The exposure estimation for the Tier 1 risk assessment considers the default RUD value of 1 mg/kg (i.e. relevant SVs are calculated by considering this RUD value). In the next steps the soil persistence of the substance as a triggering factor is considered and if further assessments were necessary, it is suggested to take into consideration the pore water concentration of the root zone of the crop when flowering and combine it with an additional factor of 10. The current EFSA GD has been criticised because almost all substances (even rather non-persistent substances and substances known to present low toxicity to bees) triggered higher tier assessments and no detailed guidance was made available about how to calculate the relevant pore water concentration. The following questions aim to explore the possibility of refining the exposure assessment for the succeeding crops scenario at lower tiers.

1. Are the "persistence in soil trigger values" of 2 days and 5 days appropriate to require/exclude the risk assessment for the succeeding crop scenario?
2. If the soil DegT50 of a substance is a valid trigger for the succeeding crop scenario, how this "persistence in soil trigger value" should be selected from the available soil DegT50 dataset?
3. Is there any other factor/parameter (or a combination of factors/parameters) contributing to the fate of PPP residues in pollen and nectar that could be used to identify a rapid and cost-effective initial assessment?
4. Is the actual methodology used to calculate the 90<sup>th</sup> percentile of the average pore water concentration in the root zone suitable?
5. Is it appropriate to derive PEC<sub>pollen</sub> and PEC<sub>nectar</sub> from the PEC<sub>pore water</sub>?

###### 4.1.1.2. Proposed approach

Question 1 and 3 above could be answered by gathering information and evidences from the available literature studies and taking into consideration the recommendations of current guidance documents in the environmental fate and behaviour and residues areas addressing the persistence in soil and the plant uptake/translocation processes. For example, a screening criterion based on the molecular weight of the compound could be considered based on the results of uptake studies with various crops/compounds combinations conduct

by Lamshoeft et al. (2018), indicating that crop uptake is negligible for compounds with a molecular weight > 394 g/mol.

It is proposed not to investigate further the “systemicity” concept as no clear physical-chemical/fate properties driven definition exist, and even if such a robust and scientifically sound methodology to characterize “non systemic” active ingredients which would not be translocated to pollen and nectar (and thus potentially not triggering an assessment for the succeeding crop scenario) was available, there are ways to render non systemic products systemic, by adding, e.g., copolymers to the pesticide formulation (e.g. Ishaque et al., 2014).

Answer to question 2 is quite straightforward based on the experience and the expert judgement in the regulatory environmental fate area and will be aligned with the current regulatory practices in that area.

Question 4: The use of the FOCUS GW scenario that is most relevant to the area of use of the substance to derive the PEC<sub>pore water</sub> can be considered superseded by the new EFSA PECsoil Guidance (EFSA, 2017) with soil exposure scenarios targeted to the concentration in the root zone. In this case the exposure is referred to the 90<sup>th</sup> percentile concentration (peak in time resulting from multiyear applications) considering all agricultural fields within a regulatory zone where a PPP is intended to be used. The output to be considered from the 4-tiers based exposure assessment is the PEC<sub>pore water</sub> in mg/L averaged over the top 20 cm of soil based on average substance properties. The limitations of the GD is that it does not cover all cropping and application systems such as drip irrigation, dipping and drenching and large (with a diameter > 0.5 cm) treated seeds application (maize seeds and pelleted seeds are considered by this GD as small seeds). In addition, the default soil layers depths may not reflect the root zone depths of the cop in question.

Question 5: With the new software tools of the EFSA soil persistence GD (the simple analytical model PERSAM and the modified versions of the pesticide fate models PEARL and PELMO), the correlation between PEC<sub>pore water</sub> and PEC<sub>pollen</sub>/PEC<sub>nectar</sub> will be further investigated.

#### 4.1.2. Weeds in field scenario

##### 4.1.2.1. Background and problem formulation

In EFSA’s 2013, exposure through pollen and nectar from flowering weeds in the treated field is one of the exposure scenarios that need to be considered in the risk assessment. However, over the past years it has been questioned whether or not this exposure scenario should be considered relevant. In Section 2.3 of Appendix N of the EFSA, 2013, the following is stated:

“If the first step results in an unacceptable risk, it may be checked whether it is likely that a significant fraction of the surface area of the treated fields is covered by weeds at the application time. If this is likely in less than 10% of the area of use of the substance, no weeds will occur in a 90th percentile case and thus their exposure can be ignored. For example, weeds are usually not abundant in annual crops - abundant weed growth is more likely to occur in, for example, orchards. However, at this moment no guidance for the assessment of the abundance of weeds is available for most crops”.

Based on the text cited above, assessing whether exposure through flowering weeds in the treated field is a relevant exposure scenario requires 2 separate steps:

1. Determine what fraction of the surface area of a single treated field has to be covered by weeds in order for this fraction to be considered 'significant'.
2. Consider all fields in the area of use of the substance, and determine in what percentage of these fields the weed coverage is higher than that 'significant fraction of the surface area'. If this is the case in less than 10% of all fields, no weeds will occur in the 90<sup>th</sup> percentile case, and thus this exposure scenario can be considered not relevant.

It should be noted that in the ESA, 2013, the 'significant fraction' from point 1 above has not been determined or specified. This is further discussed in Section 4.1.2.5.

In addition to the surface area of the field covered with weeds, the relevance of this exposure scenario also depends on the growth stage of the weeds present (i.e. only weeds at the flowering stage will be a potential source of pollen and nectar). Furthermore, the time of application (i.e. the growth stage of the crop) also plays an important role. Therefore, in order to assess the relevance of the flowering weeds scenario for the risk assessment, an answer should be provided to the following research question:

What is the distribution of occurrence (% of ground coverage) of weeds at different growth stages in permanent and arable crops, in relation to crop growth stage?

#### 4.1.2.2. Methods for data collection

To address the research question, results from studies that have investigated the occurrence of weeds in different crops are needed. The information available should include the percentage surface area of the field covered by flowering weeds, the BBCH growth stage of the weeds observed and the BBCH stage of the crop at the time of the assessment.

The most important data source are studies performed by industry, as different companies have carried out specific studies that investigated the occurrence of weeds. For example in the context of the confirmatory data request for clothianidin and imidacloprid (following Regulation (EU) No. 485/2013), two different notifiers each submitted a study addressing the presence of flowering weeds in different crops, for which the study report and raw data are available to EFSA:

1. **Maize and potato:** *5.1.2.e Woo (2014) Identification of weeds population and honeydew presence in maize and potato fields during the growing season. Report no. THW-0383. Not published*

This study is a monitoring study, in which the presence of weeds throughout the growing season was monitored on about 50 commercial fields of either maize or potato in different European countries.

2. **Cereals, sugar beet and potatoes:** *5.1.2.e Woo (2014) Evaluation of the occurrence of flowering weeds in agricultural crops: cereals, sugar beet and potatoes. Report no. M-505126-01-1. Not published*

In this study data on the occurrence of weeds from control plots of herbicide efficacy trials in cereals, sugar beets and potatoes, conducted between 2004 and 2014, were analysed.

In an attempt to more generally demonstrate that the flowering weeds scenario should not be considered relevant, the European Crop Protection Association (ECPA) launched a project to analyse the data on the presence of weeds in control plots of herbicide efficacy trials from different crops, supplied by a number of companies (Syngenta, Bayer Crop Science, BASF, Dow Agrosciences (now Corteva Agriscience) and Monsanto (now part of Bayer)). Within this project, data was available from eight arable crops (**cereals, maize,**



**oilseed rape, sunflower, potatoes, sugar beet, peas and beans**) and three permanent crops (**orchards, citrus and grapes**). This dataset includes all the data analysed in the study by [5.1.2.e Woo](#) et al. (2014). Some preliminary results were published by Maynard et al. (2014). In 2019, the finalized assessment was reported in an unpublished regulatory report by [5.1.2.e Woo](#) et al. (2019). The full study report and the complete dataset used for this assessment were made available by ECPA to the Working Group.

In summary, two main datasets are available: a first dataset from the monitoring study by [5.1.2.e Woo](#) (2014), and a second dataset with weed recordings from herbicide efficacy trials from the ECPA project ([5.1.2.e Woo](#) et al., 2019). In the context of the revision of the EFSA, 2013, both datasets were assessed in detail. As some shortcomings were identified in the analysis performed by the study authors, the datasets were re-evaluated to better address the research question.

Due to the specific nature of the data required (i.e. parameters that need to be measured) to address the research question, it is not expected that suitable data would be available from studies available from published literature. Therefore, a search of the published literature was not and will not be performed in this case.

#### **4.1.2.3. Dataset 1: monitoring study in maize and potato**

The study by [5.1.2.e Woo](#) (2014) was submitted in the EU in the context of the confirmatory data request for clothianidin following Regulation (EU) No. 485/2013). For a detailed summary of the methods and results, reference is made to the Addendum to the DAR of clothianidin (EFSA, 2016a).

##### *Methods used for obtaining the dataset*

This study was a monitoring study. The presence of weeds throughout the growing season was monitored in 2014 on 53 maize fields (spread over France, Italy and Hungary) and on 55 potato fields (spread over France, Italy, Spain, Germany, United Kingdom, Hungary and Poland). The fields where assessments were made were commercial fields in the majority of cases. Some fields for efficacy studies or registration trials were also included. As these fields were conventionally managed, herbicide applications were performed to control weeds.

In each field, 8 observation plots were selected for monitoring. For each observation plot at the field site, the number of weeds/plot was assessed by counting the weeds present over the whole plot. Each weed present was identified, so that the number of each species was recorded as well as its development stage (using the BBCH scale). The observations were carried out on three occasions: one month after sowing of the crop; at crop flowering; about mid-September.

##### *Analysis and results reported in [5.1.2.e Woo](#) (2014)*

In the results section of the study report, only the name of the weed species that were flowering at the time of assessment was reported, together with the number and percentage of sites where each respective species was found. Further, also the average density over all sites where the species was present is reported. The results of this study indicate that flowering weeds are generally only present in low numbers in maize and potato.

However, no information on the weed ground cover was available. Further, the results were presented and discussed for each weed species separately, while at most field sites more than one weed species was present at the flowering stage. Consequently, the information included in the study report provides little useful information in the context of the research question, for which information on the total ground cover of all flowering weeds present in a field is needed.

##### *Results from the re-evaluation reported in the Addendum to the DAR of clothianidin*

During Peer Review of the confirmatory data for clothianidin, the RMS was requested to re-evaluate the raw data, and to provide a rough estimation of the total weed ground cover at the field sites monitored in this study. The outcome of this re-evaluation is included in the Addendum to the DAR of clothianidin. A summary is reported below.

Based on the raw data available in the study report, the total number of weeds (for all species) counted at each observation time (1 month after sowing, at crop flowering, mid-september) was determined for each field site. A distinction was made between weeds that were not flowering (BBCH < 60) and weeds at the flowering stage (BBCH ≥ 60).

For both maize and potato, the **number of sites with weeds** (all BBCH stages) **and the number of sites with flowering weeds** (BBCH ≥ 60) was determined for each observation time. The data shows that while flowering weeds were only found on a limited number of sites one month after sowing, they in general become more abundant during the course of the growing season (e.g. for maize flowering weeds were found at 7.5% of the sites one month after sowing, and in 43% of the sites mid-September). The **total weed density per site** was also determined. The density (weeds/m<sup>2</sup>) generally increased throughout the growing season. Further, the maximum density of flowering weeds in potato is consistently lower compared to the maximum density in maize.

A direct **estimation of the percentage of the ground surface covered by flowering weeds** at the tested field sites is not possible, as no information is available in the study report on the weed ground cover.

#### *Conclusion*


The most interesting finding from the present study in maize and potato is that (flowering) weeds are found only on a limited number of sites early in the crop development, but that they in general become more abundant during the course of the growing season. As the ground coverage of the (flowering) weeds was not measured, this dataset does not provide any other useful information to address the research question outlined in Section 1.

#### **4.1.2.4. Dataset 2: weed recordings from control plots of herbicide efficacy trials**

##### *Composition of the dataset*

Data from industry herbicide efficacy trials (control data only) from different crops were made available via an ECPA working group. As efficacy trials are conducted to recognised standards and guidelines across Europe, the study authors consider these data to be robust, reliable and generated using consistent methodologies. Each of the contributing companies provided trial data from internal databases for one or more specific crop species. The different crops for which data are available and the number of trials for which data was included in the dataset is shown in Table 3.

The information provided by the different companies included the trial ID number, location (co-ordinates of the trial site, postal code, country), information on the plots (number of replicates, plot size), whether the trial was conducted to GEP, date of the trial, crop species, crop BBCH stage (min, max, majority), weed species, weed BBCH growth stage (min, max, majority), weed diameter and height, weed percentage ground cover, weed density. It should however be noted that for a large part of the trials, data was not available for one or more of the parameters listed above. For example, information on the weed BBCH stage and ground cover was only available for a small part of the trials (see Table 3).

Based on maps of the distribution of the trial locations for each crop included in the study report by  et al. (2019), the trials generally seem to be well spread over the different regulatory zones in Europe. However, trials performed in the Northern zone were a minority, and even lacking for some of the crops. Further, for some crops (e.g. citrus) the data was

limited to trials performed in Southern Europe. However, this can generally be explained by the geographical spread of the regions where the crop is typically grown.

*Analysis reported in <sup>1.2.6</sup>Wol et al. (2019) and its shortcomings to address the current research question*

In the first part of the assessment, the authors focussed on the **individual weed recordings**. A weed recording was defined as the act of positively recording the presence of a weed during an observation event. For each trial, each individual weed species recorded on a single assessment date was counted as one weed recording. Therefore, multiple rows of data exist for each trial if more than one weed species was observed and/or if more than one observation events (i.e. on multiple dates) occurred.

For each of the crop species for which data was available, it was determined how many individual weed recordings were made. In a next step, it was determined in how many of these weed recordings the measured weed species was at a growth stage of BBCH 60-69 (i.e. at a flowering growth stage). In a third step, a distinction was made between weed species that were attractive and not attractive to bees. For the purpose of their assessment, the authors assumed that monocot species are not attractive, and that dicot species are attractive to bees. Finally, the number of attractive weed species (i.e. dicots) at the relevant growth stages (BBCH 60-69) and with ground coverage > 10% were determined. This number of weed recordings was then expressed as a percentage of the total number of weed recordings per crop, thereby giving the proportions of attractive flowering weeds that were present in the crop area as well as the proportion of attractive flowering weeds that were present at >10%.

The assessment focussing on individual weed recordings does not provide any useful information to address the first research question, as this only focussed on a single weed species. Based on the criteria in Appendix N of the EFSA, 2013, the total weed ground cover in a field should be considered.

Further, some assumptions made are questionable. As acknowledged in the study report, weeds can reach the flowering growth stage quickly. Therefore, focussing only on weeds from BBCH stage 60-69 might be too limited. Although it is stated in the study report that BBCH stage  $\geq 30$ ,  $\geq 40$ ,  $\geq 40$  and  $\geq 70$  were also included in the assessment, the outcome of this assessment is not reported. Additional consideration of recordings of weeds in these other growth stages is considered necessary.

To determine whether a weed species was attractive to bees, a distinction was made between dicot and monocot species, with only dicot species considered to be attractive. As acknowledged by the study authors, there are exceptions to this rule. It is therefore considered to be a rather arbitrary way to determine the attractiveness of weeds to bees. Further, in most trials the majority of the weeds recorded were dicot species. Therefore, and in absence of more clear criteria to determine the attractiveness of a weed species, it is considered more appropriate to assume that all recorded weeds are attractive to bees when flowering.

For the last step in this assessment process, the number of recordings with a flowering weed at >10% ground coverage was compared to the total number of weed recordings to determine a percentage. However, not for all weed recordings considered, data on the ground coverage is available. Consequently, the authors assumed that for those recordings where no data on the ground coverage is available, the weed coverage was less than 10%. It is however not possible to prove that this assumption is correct based on this dataset (i.e. it might very well have been the case that although the % ground coverage was not recorded, it was > 10%). Therefore, the 'percentage of attractive weeds > 10% ground coverage' as presented in the tables in the report cannot be considered a reliable value. In addition, the value of 10% as a trigger to determine whether weed coverage within a field is significant is not supported by data (see Section 4.1.2.5).

In a second part of the assessment, the **total percentage ground coverage of weed species per trial** was determined, by adding together the percentage ground coverage values for individual weed species (attractive and flowering weeds only) within each trial. Any trial which resulted in total weed coverage values of >100% were adjusted to a value of 100% (i.e. assumed total weed coverage). Based on the determined values, mean, minimum, maximum, 90<sup>th</sup> percentile and 95<sup>th</sup> percentile values for total ground cover of flowering attractive weeds was calculated. This was again done for each of the crop species for which data was available.

As in this part of the assessment the total weed ground cover for a trial is considered, this assessment is more relevant in relation to the research question. However, it should be noted that when calculating the total percentage ground cover of weeds per trial, the authors did not take into account whether or not multiple assessments were performed within a single trial. For example, if for one trial weed recordings were available for two assessment dates, a single total percentage ground coverage for this trial was calculated, summing up all weed recordings for both assessment dates. This way, important information related to the temporal scale (when the assessment were made, how many times, etc.) was lost. In addition, the assessment did not relate the total ground cover of weeds to the crop BBCH stage.

In addition to the assessments discussed above, the **monthly distribution of weed recordings** was plotted for each crop, as was the **monthly distribution of the weed BBCH growth stage**. The latter plots indicated that, with the exception of oilseed rape, flowering weeds were generally only present in arable fields during the months of March, April, May, June and July. Based on these observations, the authors concluded that the 'flowering weeds in the treated field exposure scenario' may only be relevant for many arable crops between the months of March and July. However, the plot of the monthly distribution of weed BBCH growth stage is heavily biased by the monthly distribution of the weed recordings, which did not cover all months of the year. Therefore, any conclusion based on these plots is considered to be of limited reliability.

Finally, histograms were plotted for each crop to present the **distribution and frequency of the crop growth stage** for the trials in the dataset. These plots show that the majority of the weed recordings were made at early crop BBCH stages only. It should however be noted that these histograms were made by taking into account the complete dataset, including also those trials for which no data on weed BBCH stage or ground cover were available. Therefore, based on these plots, the crop BBCH stage cannot directly be related to the weed BBCH stage or weed ground cover.

#### *Re-evaluation of the dataset in the context of the revision of the EFSA, 2013*

As discussed above, a number of shortcomings were identified in the analysis performed and reported by [B.1.2.e Wot et al. \(2019\)](#). Therefore, in order to better address the first research question, this dataset was re-evaluated. The main goal of this re-evaluation was to relate the total weed ground cover in the trials to the crop BBCH stage, and to extend the weed BBCH stages considered beyond BBCH 60-69.

## 1. Methods

The analysis was performed for each crop species separately, as described below. As a first step, the Excel files containing the raw data for each crop were cleaned in Excel 2016, by removing all the columns that were not relevant for the present assessment. The following columns were retained: Trial ID, Application date, EPPO Crop code, Crop BBCH (min, max, majority), EPPO weed code, Weed BBCH (min, max, majority), percentage ground cover. Further cleaning operations were intended to allow for the further steps in the analysis to be performed in the R statistical environment (e.g. replacing empty cells by 'NA'). The actual manipulations of the dataset and the calculation of the total weed ground cover

and summary statistics was performed in the R statistical environment, Version 3.5.1 (R Development Core Team, 2018).

For each row (i.e. weed recording), it was checked if a value was available in the column 'crop BBCH majority' or 'weed BBCH majority'. If this was not the case, the value available from the same row in the column 'crop BBCH max' or 'weed BBCH max' was copied (if available) to the column 'crop BBCH majority' or 'weed BBCH majority', respectively<sup>1</sup>. Afterwards, all rows with blanks (i.e. no value) in either the columns 'weed BBCH majority' and 'percentage ground cover' were removed from the dataset. That way, only those trials with data on weed BBCH stage and percentage ground cover of the weeds were retained for further analysis.

As the aim of the assessment was also to link the weed ground cover with the crop BBCH stage, all rows with blanks in the column 'crop BBCH majority' were also removed, resulting in a dataset containing only trials for which crop BBCH stage, weed BBCH stage and percentage ground cover of the weeds. For the permanent crops, however, this last operation was not performed. This was because for permanent crops, for most of the trials with data on weed BBCH and ground cover, the crop BBCH stage was not measured. Given that the dataset for permanent crops was already small, considering only those trials with data on weed BBCH stage, ground cover and crop BBCH stage would result in a dataset that is too small for any meaningful analysis. Further, for permanent crops, the competition (mainly in terms of space) between the crop and weeds will be less dependent on the crop BBCH stage compared to arable crops, especially if the permanent crop is already well established. Consequently, it is considered that the occurrence of weeds in permanent crops is less dependent on the crop BBCH.

The number of trials for each crop that was retained for further analysis is shown in Table 3. In some trials, weed assessments were performed on more than one occasion (different dates). Consequently, for some of the crops, the total number of assessment occasions exceeded the total number of trials. Therefore, the analysis focused on each assessment occasion separately, rather than on each individual trial as a whole.

**Table 3. The total number of trials in the original dataset, the number of trials retained in the dataset used for the re-evaluation, the number of trials where the abundance of weeds was assessed on 1, 2, 3, 4 or 6 occasions, and the total number of assessment occasions for each crop in the dataset.**

Crop		Total number of trials in the original dataset	Number of trials retained for analysis <sup>1</sup>	Number of trials with ... assessment occasions					Total number of assessment occasions
				1	2	3	4	6	
Arable	Sunflower	390	37	34	2	1	0	0	41
	Maize	7669	2509	2047	411	43	8	0	3030
	Oilseed rape	923	38	38	0	0	0	0	38
	Cereals	982	343	229	83	24	7	0	495
	Sugar beet	140	45	6	3	27	8	1	131
	Potato	355	59	36	18	5	0	0	87
	Pea	659	148	90	43	9	6	0	227
	Bean	192	64	34	25	3	2	0	101
Permanent	Grapes	139	71	71	0	0	0	0	71
	Orchards	51	12	12	0	0	0	0	12
	Citrus	40	8	8	0	0	0	0	8

<sup>1</sup>For permanent crops only those trials with data on weed BBCH stage and percentage weed ground cover were considered, while for arable crops only trials that in addition to the above had data on crop BBCH stage were considered.

In a next step, the dataset for each crop was split into separate subsets for different groups of crop BBCH stages (i.e. 0-9, 10-19, 20-29, ...) for which data was available. For each of these crop BBCH stage subsets, the total percentage ground cover of weeds above a certain BBCH stage (i.e.  $\geq 30$ ,  $\geq 40$ ,  $\geq 50$  or  $\geq 60$ ) was calculated per assessment occasion. In those cases where the calculated total weed coverage exceeded 100%, the value was adjusted to 100% (i.e. assuming total weed coverage). Next, the number of assessment occasions for which the weed ground cover exceeded a certain percentage (i.e. 0, 5, 10, 15 or 20%) was calculated. Finally, summary statistics (min, max, mean, 90<sup>th</sup> percentile and 95<sup>th</sup> percentile) were calculated for the percentage groundcover for each combination of crop BBCH stage group and weed BBCH stage.

It should be noted that, as a conservative approach, attractiveness of a weed to bees was only considered to depend on the developmental stage of the weed (i.e. flowering or not). As discussed above, there are doubts whether the distinction dicotyl – monocotyl is acceptable as criterion to distinguish between flowering weeds that are attractive to bees or not. In absence of another clear criterion, it was considered for the purpose of this analysis that all flowering weeds are attractive to bees.

As also discussed under above, weeds can reach the flowering growth stage quickly. Therefore, the assessment was done not only taking into account weeds of BBCH stage  $\geq 60$  (i.e. weeds that are or have recently been flowering), but also weeds of BBCH stage  $\geq 30$ ,  $\geq 40$  or  $\geq 50$ , to also account for weeds that might soon be flowering.

## 2. Results and discussion

The results of the re-evaluation are summarized below for arable and permanent crops. The detailed results for the maize dataset are shown in Table 4, as an example. For the other crops in the database, similar tables were constructed, but these are not reported here. All tables will probably be included in the revised guidance document.

### **Arable crops**

For the lowest **crop BBCH stages (0-9)**, in general the number (and percentage) of assessment occasions where weeds were found are low. Only in maize (93 out of 675 assessments) and sunflower (1 out of 7 assessments) weeds of BBCH  $\geq 30$  were present (ground cover  $> 0\%$ ). Weeds of BBCH  $\geq 60$  were only recorded in maize, but only in 29 out of 675 assessments (4.3%). Consequently, if the presence of flowering weeds at  $> 0\%$  ground cover would already be considered significant, this significant fraction would only be reached in less than 10% of the fields. Therefore, exposure through flowering weeds seems to be negligible for crops at BBCH stages 0-9.

For **crop BBCH stages between 10 and 19**, weeds were observed in all crops with the exception of cereals. The number (and percentage) of assessment occasions where weeds of BBCH  $\geq 30$  were found (ground cover  $> 0\%$ ) remain however relatively low, ranging from 8 out of 162 assessments (4.9%) in peas up to 539 out of 2177 assessments (25%) in maize. Weeds at BBCH  $\geq 60$  were, however, only found (ground cover  $> 0\%$ ) in maize (149 out of 2177 assessments – 6.8%), beans (4 out of 64 assessments – 6.25%) and peas (1 out of 162 assessments – 0.6%). Consequently, if the presence of flowering weeds at  $> 0\%$  ground cover would already be considered significant, this significant fraction would only be reached in less than 10% of the fields. Therefore, exposure through flowering weeds seems also for crops at BBCH stages 10-19 to be negligible.

For **crop BBCH stages between 20 and 29**, no trials with sugar beet were available. For the other arable crops, the number of assessments performed at these BBCH stages was also considerably lower compared to crop BBCH stages between 10 and 19. Compared to crop BBCH stages between 0 and 19, in all crop species weeds are found more frequently when the crop was at BBCH 20-29. The assessment occasions where weeds of BBCH  $\geq 30$

were found (ground cover > 0%) ranged from 2 out of 18 assessments (11%) in peas to 97 out of 170 assessments (57%) in maize. Note that for oilseed rape (0 out of 2 assessments) and sunflower (3 out of 3 assessments) more extreme values were found, but for these crops the number of assessments is considered too low for a meaningful conclusion. Weeds at BBCH  $\geq 60$  were only found (ground cover > 0%) in cereals (12 out of 142 assessments – 8.5%) and maize (9 out of 170 assessments – 5.3%). Consequently, if the presence of flowering weeds at > 0% ground cover would already be considered significant, this significant fraction would only be reached in less than 10% of the fields. Note that for sunflower, in 1 out of 3 assessments weeds at BBCH  $\geq 60$  were found, but as already mentioned above the number of assessments for this crop is too low for a meaningful conclusion. Therefore, exposure through flowering weeds seems also for crops at BBCH stages 20-29 to be negligible.

For **crop BBCH stages between 30 and 39**, again a lower number of assessments is available. With the exception of maize, the number of assessments where weeds are found at these crop BBCH stages is again higher compared to crop BBCH stages between 20 and 29. Not taking into account sunflower and oilseed rape (for which only 1 trial is available at crop BBCH 30-39), the assessment occasions where weeds of BBCH  $\geq 30$  were found (ground cover > 0%) ranged from 2 out of 8 assessments (25%) in maize to 39 out of 51 (76%) in cereals and 6 out of 6 (100%) in beans. For the crops for which data was available, weeds at BBCH  $\geq 60$  were not found in potatoes, peas and beans. In cereals and maize, weeds at BBCH  $\geq 60$  were found (ground cover > 0%) in 11 out of 51 (22%) and 1 out of 8 (12.5%) assessments, respectively. For cereals, weeds at BBCH  $\geq 60$  were found at > 5 % ground cover in 7 out of 51 assessments (14%), and at > 10% ground cover in 4 out of 51 assessments (7.8%). Overall, whether exposure through flowering weeds can be considered negligible for crops at BBCH stages 30-39 seems to depend on the threshold for the fraction of the field surface that is considered significant.

Assessments performed at **crop BBCH stages  $\geq 40$**  are only available for sunflower, oilseed rape, potatoes, peas and beans, but their number is too low to be able to draw a reliable conclusion regarding the occurrence of flowering weeds.

#### **Permanent crops**

For permanent crops, the available data shows that weeds are in general highly abundant. When considering all weeds at BBCH  $\geq 30$ , weeds were present (ground cover > 0%) on about 75% of the assessment occasions. Focusing only on weeds at BBCH  $\geq 60$ , weeds were present (ground cover > 0%) on about 40% of the assessment occasions, and exceeded 20% ground cover on 8 to 38% of the assessment occasions. Consequently, even if it would be assumed that 20% of the surface area of the field would have to be covered for flowering weeds to be a significant route of exposure, this ground coverage would be reached in more than 10% of the fields. Therefore, the flowering weed scenario will be significant in the 90<sup>th</sup> percentile case.

**Table 4. Maize – Number of assessment occasions for which the weed ground cover exceeded a certain percentage and summary statistics of the results for percentage ground cover for weeds of a BBCH stage above a certain value, for the different growth stages of the crop for which weed recordings were performed**

Crop BBCH	Total Number of assessments <sup>1</sup>	Weed BBCH	Number of assessments with total ground cover of ...					Percentage ground cover <sup>2</sup>				
			> 0%	$\geq 5\%$	$\geq 10\%$	$\geq 15\%$	$\geq 20\%$	Min	Max	Mean	90 <sup>th</sup> %ile	95 <sup>th</sup> %ile
0-9	675	$\geq 30$	93	80	67	61	52	0.00	100	4.65	7.60	30.0
		$\geq 40$	51	41	28	28	21	0.00	100	2.31	0.00	10.0
		$\geq 50$	43	33	20	20	17	0.00	100	2.00	0.00	3.30
		$\geq 60$	29	21	13	13	12	0.00	100	1.22	0.00	0.00
10-19	2177	$\geq 30$	539	454	381	313	264	0.00	100	7.31	25.0	50.0

		≥ 40	325	252	193	157	132	0.00	100	3.49	5.00	21.2
		≥ 50	285	215	161	130	110	0.00	100	2.83	4.00	20.0
		≥ 60	<b>149</b>	<b>94</b>	<b>65</b>	<b>46</b>	<b>38</b>	<b>0.00</b>	<b>100</b>	<b>1.14</b>	<b>0.00</b>	<b>3.00</b>
20-29	170	≥ 30	97	97	91	76	72	0.00	100	30.9	100	100
		≥ 40	27	26	23	19	19	0.00	100	7.52	25.0	75.5
		≥ 50	14	14	12	9	9	0.00	100	3.22	0.00	18.2
		≥ 60	<b>9</b>	<b>9</b>	<b>8</b>	<b>6</b>	<b>6</b>	<b>0.00</b>	<b>100</b>	<b>2.53</b>	<b>0.00</b>	<b>2.75</b>
30-39	8	≥ 30	2	2	2	2	2	0.00	100	15.9	48.9	74.5
		≥ 40	2	2	2	2	2	0.00	100	15.9	48.9	74.5
		≥ 50	2	2	2	2	2	0.00	100	15.9	48.9	74.5
		≥ 60	<b>1</b>	<b>1</b>	<b>1</b>	<b>1</b>	<b>1</b>	<b>0.00</b>	<b>100</b>	<b>12.5</b>	<b>30</b>	<b>65.0</b>

<sup>1</sup>The total number of assessments that took place when the crop was at the respective BBCH growth stage. As for some of the trials an assessment was performed at more than one occasion, the total number of assessments considering all growth stages (3030) is higher than the total number of trials (2509); <sup>2</sup>Summary statistics were calculated taking into account all assessments at the respective crop BBCH growth stage (also those where weeds of the respective BBCH stages were not present).

### 3. Conclusions

This dataset contains useful information on the occurrence of weeds in different arable and permanent crops. Although only a portion of the trials included in the dataset contained the necessary information on weeds BBCH stage, weed ground cover and crop BBCH stage, this portion is still considered sufficiently extensive.

As some shortcomings were identified in the analysis performed by [S.1.2.e Wc](#) et al. (2019), a re-evaluation was performed in order to better address the first research question. Based on the results of this re-evaluation, the presence of flowering weeds in the **arable crop species** for which data is available could be considered negligible in the 90<sup>th</sup> percentile case for crops at BBCH stages 0 to 29, even if the occurrence of flowering weeds at >0% ground cover is considered as significant. For crop BBCH stages 30-39, the conclusion on relevance of the flowering weeds scenario seems to depend on the fraction of the field surface covered by weeds that is considered as significant. Due to the lack of assessments made at crop BBCH stages ≥ 40, no conclusions can be drawn for these crop BBCH stages.

Although the dataset for permanent crops is more limited in terms of number of trials, the results of the re-evaluation indicate that (flowering) weeds are in general highly abundant in these crops. Therefore, the flowering weed scenario seems to be a significant exposure scenario in permanent crops.

#### 4.1.2.5. The 'significant fraction' of surface area covered by weeds

As discussed in section 4.1.2.1, the first step in assessing whether exposure through flowering weeds is a relevant scenario comprises of determining what fraction of the surface area of a single field has to be covered by weeds in order for this fraction to be considered as 'significant'. However, in the EFSA, 2013, it is not further specified what that 'significant fraction' of the surface area of a field covered by weeds should be, or how it should be determined.

Over the past few years, industry has made attempts to demonstrate that the abundance of flowering weeds should be considered negligible, e.g. in the context of the confirmatory data for the neonicotinoids or in a recent report by [S.1.2.e Wc](#) et al. (2019) (see also previous sub-sections). In their argumentation, they consistently use a threshold of 10% weed ground cover within a single field, referring to Appendix N of the EFSA, 2013. As this threshold of 10% refers to the weed ground cover at field scale, this refers to the 'significant fraction' within a field. Specific data or an argumentation to underpin the assumption that a weed ground cover within a field of below 10% is not significant for bees has not been provided. It



is therefore assumed that this threshold of 10% originates from a misinterpretation of the text in Appendix N of the EFSA, 2013.

At present, the WG is not aware of any data that could help to determine the exact threshold above which the fraction of the surface area of a field covered by flowering weeds becomes 'significant'. It is however likely that a fraction of 10% might already result in a significant exposure in some cases.

Honey bees are known to use a collective patch-based foraging strategy, and focus their forager force on a few high-quality patches within its foraging area (Visscher and Seeley, 1982). This would imply that flowering weeds in a field will only be visited if their abundance is sufficiently high. However, some solitary bee species (i.e. oligolectic species) are specialized to feed on plants of one genus or one family (Michener, 2007; Rollin et al., 2013; Westrich, 2018). If plants of that genus are present in a treated field, it is likely they will be extensively visited by these bee species (Forrest et al., 2015). Consequently, this exposure route will be relevant for these species, even if the surface area of the field covered is below 10%. The threshold for the 'significant fraction' thus seems to depend on the feeding habits of different bees.

Further, considering only the surface area of a field in terms of percentage of the whole surface does not take into account a potential edge effect. As discussed in Section 2.8.2 of the EFSA Scientific Opinion for non-target terrestrial plants (EFSA PPR Panel, 2014), arable weeds are generally more common at the edge of field and field corners. If a high number of weeds is present in the edge of the treated field, it is well possible that these weeds become an important food source for bees living close to the field, even though the average percentage of weed ground cover relative to the whole field surface is lower than 10%. This might especially be relevant for large agricultural fields. Therefore, the threshold for the 'significant fraction' also depends on the size of the field.

On the other hand, it is reasonable to assume that the presence of a single or a few weed plants in an agricultural field will not immediately result in a relevant exposure to bees. Therefore, the threshold for a significant fraction of weed ground cover will be above 0%.

Based on the above, it is not possible to determine the exact threshold below which the percentage weed ground cover within a field is to be considered as not significant. However, it is considered reasonable to assume that it will be somewhere between 0 and 10%.

#### 4.1.2.6. Preliminary, overall conclusions

##### *Arable crops*

Both the dataset from the monitoring study by [5.1.2.e Woo](#) (2014) and from the control plots from herbicide efficacy trials by [5.1.2.e Woo](#) et al. (2019) are considered to contain reliable data on the presence of weeds in agricultural fields throughout Europe. Both datasets indicate that the presence of weeds is low when the crop is at an early growth stage. However, they also show that with increasing crop growth stages, the presence of (flowering) weeds also increases.

Based on the re-evaluation of the data from the herbicide efficacy trials, the percentage of assessments where flowering weeds (i.e. weeds at BBCH  $\geq 60$ ) were found at  $> 0\%$  ground cover was below 10% for crops at BBCH stages from 0 to 29. Thus, even if just the presence of a single flowering weed would be considered as significant, this would occur in less than 10% of the cases. Therefore, no flowering weeds will be present in the 90<sup>th</sup> percentile case. In conclusion, **for crops at BBCH stage 0-29, exposure through pollen and nectar of flowering weeds is not considered a relevant scenario, at least for the contact risk assessment.**

Data is available for 8 arable crops (i.e. sunflower, maize, oilseed rape, cereals, sugar beet, potato, pea and bean). Although the crops for which data is available are variable in terms of

plant morphology and agricultural practices, there might be some uncertainty when extrapolating the results to other crops, such as fruiting vegetables or leafy crops. However, given that the results for all 8 crops in the database are comparable for BBCH 0-29, it is considered reasonable to assume that the conclusions for crop BBCH 0 - 29 apply to all arable crops.

For crops at BBCH stage 30-39, flowering weeds were not found in 3 crops (potatoes, peas and beans), while they were present at > 0% ground cover in more than 10% of the fields in cereals and maize. Note that for the other crops, no trials performed at these crop BBCH stages were available. For cereals, weeds at BBCH  $\geq$  60 were found at >5% ground cover in 14% of the assessments, and at >10% ground cover in 7.8% of the assessments. Overall, at these crop BBCH stages, whether exposure through flowering weeds can be considered negligible seems to depend on the fraction of the field surface that is considered significant.

As discussed above, this 'significant fraction' is likely to be below 10% (especially for some solitary bee species). Further, the number of trials in the dataset with crops at BBCH stage 30-39 is relatively limited (< 11 assessments per crop, except for cereals for which 51 assessments are available). Taking this into account, the WG is of the opinion that as a more conservative approach, a decision for the crop BBCH stages 30-39 should be based on a threshold for the 'significant fraction' between 0 and 5% rather than between 0 and 10%. This would also cover the uncertainty for extrapolation to all those crops for which no data is available at this BBCH stage. For cereals, which is a widespread crop in the EU, and also the crop with the most extensive dataset at crop BBCH 30-39, already in 14% of the assessments the weed ground cover was above 5%. Therefore, exposure to flowering weeds for crops at BBCH 30-39 cannot be considered negligible in the 90<sup>th</sup> percentile case. **Exposure to pollen and nectar from flowering weeds will be considered as a relevant exposure scenario for crops at BBCH 30-39.** This conclusion might be revised when additional data becomes available.

Assessments at crop BBCH stages  $\geq$  40 were only performed in a very limited number of trials. Therefore, no reliable conclusion on the occurrence of flowering weeds at these crop BBCH stages can be drawn based on the data available. Taking into account that both datasets indicate that the occurrence of weeds increases throughout the crop development, the conclusions from the lower crop BBCH stages cannot be extrapolated to higher BBCH stages. **Therefore, exposure to pollen and nectar from flowering weeds will be considered as a relevant exposure scenario for crops at BBCH  $\geq$  40.** This conclusion might be revised when additional data becomes available.

The fact that flowering weeds cannot be ruled out as a relevant scenario for crop BBCH stages later than 30, also implies that this scenario might still be relevant for **oral exposure** for applications made at crop BBCH 0-29. Weeds that were not flowering at the time of application might flower later on in the crop development, and contain residues of the applied product in their pollen and nectar. In addition, new weeds can emerge and flower after application, and could potentially contain residues of the applied product in their pollen and nectar after uptake of residues from soil. It is acknowledged that these residues in pollen and nectar will be lower compared to pollen and nectar that were directly over sprayed, due to the different processes that are discussed in the section on the pre-flowering factor (PFF) (section 4.1.3). **Based on the outcome of the WoE exercise for the PFF, it will be decided whether or not the flowering weeds scenario should still be considered relevant for the oral risk assessment for applications made at crop BBCH 0-29.**

It should be noted that following the evaluation of the confirmatory data for clothianidin and imidacloprid (EFSA, 2016a, b), the flowering weeds scenario was not considered relevant based on the studies by [5.1.2.e Woo](#) (2014) and [5.1.2.e Woo](#) et al. (2014) for maize, potato, cereals and sugar beet. However, this conclusion was reached based on 10% as the trigger for a 'significant fraction' of ground cover within a field. As discussed above, there is no clear

scientific basis for this trigger of 10%. Further, although the lack of data for later crop BBCH growth stages was acknowledged in EFSA (EFSA, 2016a, b), there has not been a detailed assessment of the BBCH stages for which data was available, as was done in the context of the current re-evaluation of the dataset from [5.1.2.a Work et al. \(2019\)](#).

#### *Permanent crops*

In contrast to arable crops, the number of trials available in the dataset from the herbicide efficacy trials is relatively small. Nevertheless, the available data shows that weeds are in general highly abundant. When considering only weeds at BBCH  $\geq 60$ , weeds were present (ground cover  $> 0\%$ ) on about 40% of the assessment occasions, and exceeded 20% ground cover on 8 to 38% of the assessment occasions. Consequently, even if it would be assumed that 20% of the surface area of the field would have to be covered for flowering weeds to be a significant route of exposure, this ground coverage would be reached in more than 10% of the fields. Therefore, the data shows that the presence of flowering weeds will be significant in the 90<sup>th</sup> percentile case. Consequently, **exposure through pollen and nectar of flowering weeds is considered a relevant exposure scenario for permanent crops (all BBCH stages), for both the contact and oral risk assessment.**

As the number of trials for permanent crops available is relatively small, it might be questioned whether these data can be considered representative for the whole of Europe and for other crops than grapes, orchards and citrus. However, the WG considers that it is highly likely that data from additional trials would confirm the results of the available data.

### 4.1.3. Pre-flowering factor

#### 4.1.3.1. Background of the issue

In the current version of the guidance document (EFSA, 2013) the RUD values used for SV calculations can be grouped into three categories when the plant phenology is considered:



1. The default RUD of 1 mg/kg. This value is used for pre-emergence pesticide applications (i.e. BBCH  $< 10$ ) and for the succeeding crop scenario.
2. The default sets of RUD values as reported in Appendix F of the guidance document (based on Table F1). These sets of values are used for situations when the pesticide application is made during the flowering or made before the flowering but after the crop/plant emergence (i.e. BBCH 10-69).
3. The default SV of 0  $\mu\text{g}/\text{bee}/\text{day}$  that implicitly considers RUD of 0 mg/kg. This value is used for situations when the pesticide application is made for those phenological stages when exposure of pollen and nectar can be excluded (e.g. the treated crop scenario after the flowering, BBCH  $\geq 70$ ).

All considerations below are referring exclusively to the second category of pesticide applications (i.e. BBCH 10-69).

In practice, a very large proportion of the pesticide use falls in this second category. Within that category a very large proportion of the pesticide applications, especially spray applications, are performed before the flowering (for field crops, practically all herbicide applications, large proportion of the fungicide and insecticide applications). The Tier 1 risk assessment of the current guidance document does not allow to separate exposure estimations for pesticide applications between BBCH 10-59 (before flowering) and pesticide applications for BBCH 60-69 (during the flowering). This is because pesticide residue data for

pollen and nectar were available only for situations when the pesticide application was made during the flowering (Table F1 of EFSA, 2013). Residue data reflecting pre-flowering situations were very scarce. The consequence is that the available SVs were derived considering RUD values from trials when the pesticide application was made during the flowering and their SVs are used also for the pre-flowering situations. However, the mechanisms that led to the contamination of the nectar and the pollen are substantially different for the phenological stages with open flowers when compared to before flowering situations, and the current approach might be considered as overly conservative especially for early crop stages (e.g. BBCH 10-20) of field crops (see also table 5, below).

**Table 5. The mechanism of the contamination of nectar and pollen by spray application of pesticides in BBCH stages between 10-69 and their consideration in EFSA, 2013**

Crop stage	During the flowering, BBCH 60-69	Before the flowering period, BBCH 10-59
Illustrative figure (example)	 <p>Spray application during the flowering</p>	 <p>Spray application in early crop stages</p>
Mechanism leading to pollen/nectar contamination	The spray liquid directly deposits to the anther and to the pollen and may directly contaminate the nectary.	The spray liquid does not contaminate the pollen and the nectar directly. The pollen and the nectar may be contaminated indirectly via a series of processes: 1) a proportion of the sprayed mass contaminates the plant surface (e.g. leaves) → infiltrates into the plant → becomes mobile and distributes within the plant → reaches the reproductive organ(s) → excreted into pollen and nectar; 2) another proportion of the sprayed mass contaminates the soil → distributes in the soil profile → taken up by the roots of the plant → distributes within the plant → reaches the reproductive organ(s) → excreted into pollen and nectar.
Time to contamination	The contamination of pollen and nectar is considered as immediate.	The time between the pesticide application and the actual contamination may be considerable (e.g. 3-4 month between an early post-emergent spray application and the beginning of the flowering for sunflower or maize). During this time, some significant dissipation (including degradation) of the pesticide molecule may take place.
Exposure estimation as of EFSA, 2013	The exposure of the pollen and nectar is calculated by the guidance document considering the full	As described in the left-hand box.

	application rate (full mass sprayed) for the treated crop scenario (and for the succeeding crop scenario). For the weed scenario and off-field scenarios the application rate corrected with the Ef factor is considered (see section 3.3.2). SVs calculated by considering Appendix F of the guidance document.	
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#### 4.1.3.2. Proposed approach

In order to derive solid pre-flowering factors (which should be different for different BBCH stages), a set of residue trials should be available where the residue levels from pre-flowering applications could be compared with residue levels from during flowering applications. Alternatively, precise measurements of the different steps of the mechanism outlined above should be available. Unfortunately, these data are scarce (the dossier of imidacloprid contains some trials where the distribution of the radiolabelled molecule was followed after pre-flowering applications; recent studies of sulfoxafloflor investigated pollen and nectar residue levels from pre-flowering applications in a number of crops). Therefore, a narrative review will be conducted by the WG that will consider relevant information about the dissipation and other relevant processes of the mechanism as described above. The information that will be gathered will be weighted for their relevance to inform one or the other processes under discussion. Pending on the usefulness and relevance of the data, a proposal will be made for a set of BBCH dependent pre-flowering factors. As for example, the following data may be considered: Fantke et al, 2013; Fantke et al, 2014; Lewis et al, 2017; EFSA, 2017; Ebeling et al, 2018 .

#### 4.1.4. Protein content of pollen

##### 4.1.4.1. Background of the issue

As regards nectar, the guidance document considers that its consumption by bees depends on its quality, the higher the sugar content is the lower the consumption.

A similar concept is not considered for pollen. Currently, the guidance considers the highest pollen consumption from the pollen consumption data that was gathered that time, but Appendix S is offering the option to take into consideration the pollen quality as a refinement for pollen consumption for Tier 2. Based on the information summarized in that appendix, protein content seemed to be the driving factor, although it was not excluded that other components of the pollen played also a considerable role. The WG has considered whether such a concept (the quality/nutritional value of the pollen influences the pollen consumption) could be introduced to Tier 1 of the reviewed guidance document.

##### 4.1.4.2. Review process that was followed

A narrative review of the issue was conducted. Several publications from the open literature had been reviewed, among others, such papers as Pamminger et al., 2019; Basualdo et al., 2013; Kleinschmidt and Kondos, 1978; Zheng et al., 2014; Corby-Harris et al., 2018; Vaudo et al, 2016.

The review of these data indicated that: 1) protein content of the different crop and plant species are indeed notably different; 2) the pollen consumption and the performance of honey bee colonies fed with pollen with different origin/composition is notably different, 3) the pollen consumption was indeed different in relation to the crude

protein content according to some authors while in other papers this phenomena was not that evident (in a paper it was suggested that the level of need for brood care may influence this phenomenon); again others suggested that the crude protein content has a rather narrow optimal range suggesting a non-linear relationship between protein and pollen consumption; 4) food consumption and some physiological parameters of the bees may change significantly depending on the protein - lipid ratio; 5) the protein - hydrocarbon ratio may also play a role.

Overall, this narrative review revealed that there are some uncertainties on whether protein content alone is a good indicator for pollen quality and whether the relationship between pollen consumption and protein content is linear.

#### **4.1.4.3. Preliminary conclusions**

Considering the uncertainties as described above, the foreseen complexity of this issue and that fact that issue was considered as low priority, the WG decided not to investigate this issue further. That led to the conclusion that the Tier 1 model (see in 3.3.2) as well as the planned data collection (see section 5.2) will focus on direct pollen consumption by the bees (i.e. not protein consumption for example). However, it is not excluded that it will be recommended that the issue can be addressed at Tier 2 level risk assessment (as the current guidance document also recommends this).

#### **4.1.5. Landscape dilution factor**

##### **4.1.5.1. Background of the issue**

The exposure assessment goal of the current guidance document is linked to the concentration of the residues entering the hive/nest. It is planned that the reviewed guidance document will use the updated residue data base as described in section 5.4. This data base includes data on residues measured directly from the flowers in the field and includes data on residues entering the hive. If only the latter will be considered for the estimation of the residue intake (i.e. for the  $SV_{\text{flower}}$ ) and it is from studies where bees were allowed to forage on the landscape, than the effects of the landscape on the residue levels entering the hive will implicitly be included in the exposure estimation. If the former data will be considered (perhaps together with data from semi-field tunnel studies), this will not be the case.

However, bees forage from a diversity of flower sources including crop and non-crop flowers (Leonhardt and Blüthgen, 2012; Botías et al., 2015; Böhme et al., 2018b; Persson et al., 2018). The Tier 1 scenarios as they are in the current guidance document assume that all the food collected by bees originate from the treated field or contaminated areas around the field which is near to the hive/nest and ignores alternative food sources. Combining this with the residue data originating directly from the flower would result in an assumption that the pesticide concentration in pollen and nectar brought back to the hive/nest is the same as in the pollen and nectar in the contaminated field. The fact that the potential dilution of pesticide concentration entering the hive/nest by the collected alternative food sources (that are normally available in the landscape) is not considered would make the exposure estimation unnecessary conservative.

Appendix E in the EFSA, 2013 provides a simple model for calculating the (honeybee) hive pesticide exposure, as a step towards developing a landscape-level approach to exposure assessment. This appendix also details the alternative to use field exposure studies to introduce such landscape dilution for nectar/pollen. The guidance document proposes that a dilution factor, accounting for the diverse diet of bees resulting in reduced pesticide

exposure from a single source, should be introduced to make the risk assessment more realistic. This factor is hereafter referred to as the landscape dilution factor (LDF). LDF can be calculated as the residue concentration of a specific compound in nectar or pollen carried by forager bees at the hive/nest in relation to the concentration of that same compound in nectar or pollen from the contaminated plant, e.g. the treated crop itself (EFSA, 2013; Kyriakopoulou et al., 2017). An LDF of 1 for the treated crop scenario would mean that 100% of the food entering the hive/nest originate from treated fields.

The LDF concept might be applicable for all the scenarios for the dietary exposure. Nevertheless, this chapter will focus on the treated crop scenario only. Careful considerations will be made by the WG at a later stage to investigate whether the concept can be extrapolated to the other scenarios, and if yes, how to do this.

The extent of landscape dilution is most likely depending on the food type (pollen or nectar), identity of the treated crop (and its attractiveness), land use in the landscape, alternative flower resources and the bee group in focus. For example, bee species with short foraging ranges, foraging close to the hive/nest, may indeed focus on one type of food source in a monocultural landscape. Some solitary bee species specialized their pollen collection to certain crop species (which may suggest an LDF of 1), while social bees such as honeybees and bumblebees are (with few exceptions) generalist pollen collectors where a LDF <1 is plausible (Leonhardt and Blüthgen, 2012; Sponsler et al., 2019). Pesticide exposure to bees visiting apple orchards has been shown to be modified by natural habitat in the surrounding landscape (Park et al., 2015). In addition, landscape level pesticide exposure may be related to crops unattractive to bees through weed, drift to neighbouring plants or succeeding crops (Simon-Delso et al., 2017).

The following questions could be raised:

- 1) What is the ratio between the concentration of a compound in nectar collected from the crop field and that of the nectar brought back to the hive/nest?
- 2) What is the ratio between the concentration of a compound in pollen collected from the crop field and that of the pollen brought back to the hive/nest?
- 3) To what extent does pollen entering the hive/nest originates from a single crop or crops in general?
- 4) To what extent does nectar entering the hive/nest originates from a single crop or crops in general?
- 5) Is the landscape dilution factor as potentially derived by answering the questions in 1-4 dependent of the focal crop (and its attractiveness)?
- 6) Is the landscape dilution factor as potentially derived by answering the questions in 1-4 landscape context dependent, i.e. depending on the land use such as the proportion of crop and the availability of alternative flower resources?
- 7) Is the landscape dilution factor as potentially derived by answering the questions in 1-4 bee species/bee group dependent?

#### **4.1.5.2. Proposed approach**

The most suitable data for informing on the landscape dilution is simultaneous collection of the following after a known application of a particular compound:

- a. concentration of the compound in nectar/pollen from the treated crop (nearby the bee hives/nests)

- b. concentration of the compound in nectar/pollen brought back by bees to hives/nests located nearby to the treated crop field
- c. an alternative to a. could be concentration in nectar/pollen collected from bees foraging in the focal field (i.e. crop content analysis of bees intensively foraging within the treated field)
- d. landscape cover/land use and flower resources
- e. proportion of pollen identified to the crop plant species of all the pollen entering the hive
- f. proportion of nectar identified to the crop plant species of all the nectar entering the hive

Unfortunately, data as described in a. to c. with or without information as described in d. are scarce. Such data (a. to c.) from samples not in relation to a known application could also be informative, but less so.

The WG considered that data as described in f. is also unlikely to be available or scarce.

However, data as described in e. is available and was considered as a potential source for informing LDF (i.e. proxy for LDPpollen). Similarly, to the above, such data from samples not in relation to a landscape with known crop dominancy could also be informative, but less so.

Sources of data/information should be palynology data from dossier studies and from scientific and grey literature. Some relevant projects on pollen diversity and bee visits on plants are ongoing (INSIGNIA, C.S.I. Pollen, UrbanBee, FlorAbeilles, PoshBee). These projects could be investigated for suitability and the relevant data to be analysed for this specific purpose.

#### 4.1.6. Exposure via water consumption

##### 4.1.6.1. Background of the issue

Here we review the relevance of the water scenario to the risk assessment of plant protection products on bees. The EFSA (2013) guidance document describes the exposure via the water scenario as applying to consumption by both adult and larval bees, leading to a risk of oral exposure from three possible sources; guttation fluid, surface water, and puddles which are dealt with in section 3.5 of the EFSA (2013) guidance document. Of the three scenarios, the collection of water from guttation fluid is considered the highest risk, and is assessed as being "*based on several worst-case assumptions such as the highest water consumption rate observed in literature at 35°C (Free and Spencer-Booth, 1958) and maximum water solubility as the concentration in guttation droplets*". Whilst the current document states that water exposure should only be assessed for honey bees, the exposure for bumblebees and solitary bees is implicitly covered in this guidance as "*the very high level of water fluxes in honey bees at the colony level should be sufficiently protective for bumble bees and solitary bees*".

##### 4.1.6.2. Proposed approach

In order to investigate the relevance of the exposure via consumption of contaminated water it is necessary to understand how much of the collected water the bees consume, from which sources this water comes and when. The two general questions and related sub-questions to answer are reported in table 6.



**Table 6. General questions and related sub-question for addressing the relevance of the pesticide exposure via water consumption**

<p>Q1 What is the water consumption of adult honey bees and larvae?</p>	<p>Q1.1. How does the total water demand of bee colonies relate to water consumption only?</p> <p>Q1.2. What different sources cover the water needs of adult bees? Can this be quantified? Does the distribution between the different sources vary during the day and during the season, and can this be quantified?</p> <p>Q1.3. Same question as 1.2 for larvae.</p> <p>Q1.4. How many days of the year adult honey bees or larvae consume water collected by the water foraging bees (i.e. not only from the water contained in nectar or honey)? Which conditions (both environmental and bee behavioural) determine whether water is collected?</p>
<p>Q2 Is the exposure to pesticides by collected water (esp. guttation water) a relevant scenario?</p>	<p>Q2.1 Adult honey bees: what is the proportion of the daily pesticide intake via consumption of collected water compared to the daily pesticide intake via nectar and pollen?</p> <p>Q2.2 Larvae: what is the proportion of the daily pesticide intake via consumption of collected water compared to the daily pesticide intake via (royal or worker) jelly, nectar and pollen?</p> <p>Q2.3 Can the occurrence of guttation be characterised over the day? This is expected to be a function of (i) crop type, (ii) crop and BBCH crop development stage, and (iii) environmental conditions</p> <p>Q2.4 Can the concentration of pesticides in the guttation water be quantified as a function of time after application or sowing? This is expected to be a function of (i) compound, (ii) BBCH crop development stage, (iii) application technique (seed treatment or spray-downward or up- and sideward) and (iv) application timing</p> <p>Q2.5 How do the pesticide concentrations in guttation water, puddle water and surface water compare in general?</p>

These questions will be first considered for honey bees, thereafter information on also solitary bees and bumble bees may be gathered.

The proposed approach to answer the questions is outlined below.

## **Q1. What is the water consumption of adult honey bees and larvae?**

### **Q1.1. How does the total water demand of bee colonies relate to water consumption only?**

The collection of water by honeybees is well documented, and there is strong evidence that bees will collect and use foraged water for cooling the hive (See Seeley 1995 for a review of the literature); however, the evidence that bees will consume water to maintain their osmotic balance frequently and in large amounts is less strong.

Bees generally cover their water requirements from water present in nectar (containing approximately 80% water) and honey (18-20% water). Only when their metabolic water needs cannot be covered by consumption of nectar or honey they will need additional water. Additional water may especially be needed when: (i) there are few floral resources and nectar is scarce, or (ii) when poor weather restricts foraging, e.g. when the temperature is below 9°C, or when rain prevents foragers from flying. When no or insufficient nectar is brought in the hive the bees still need to cover their energetic demand and this will be done by either consuming freshly stored nectar or ripened honey. This implies that in all cases a considerable part of the water needs of bees is covered by intake of nectar or honey. Thus collected water always covers only part of the metabolic water need of the bees.

In addition to their metabolic water need, bees also collect water for other purposes e.g. to cool the hive, or to regulate the humidity in the hive. In general water collection for hive cooling happens only during hot days in summer, while the collection of water to cover the bees metabolic need is more important in spring and autumn.

#### **Cooling:**

If fanning is insufficient to cool a nest then bees will start to spread water, especially over the brood nest by regurgitating water droplets and covering the combs with a thin water film, to facilitate evaporative cooling. Additionally, water may be evaporated from workers tongues, as they coat their tongue in a thin layer of water that rapidly evaporates across the surface area of the fully extended tongue (Seeley, 1995). The WG questions the likelihood of non-permanent sources (puddles and guttation fluid) in contributing to cooling water under conditions leading to in-hive  $T > 33-36^{\circ}\text{C}$ .

#### **Regulating humidity**

The absolute and relative humidity of the brood chambers is highly stable in honeybees and stingless bees, whilst the humidity in the nectar stores, or in empty colonies is more variable (Human et al., 2006; Ayton et al., 2016) indicating that bees are actively regulating the humidity of the brood chamber. Maintaining a constant humidity in areas where there are brood may be the reason why water is collected when brood are present – it may not be to dilute their food.

The WG will perform an extensive literature review to estimate the relative importance of each water demand. Presently the WG tends to consider water not consumed and metabolised by bees or larvae as not relevant for the oral exposure to pesticides.

## **Q1.2. What different sources cover the water needs of adult bees? Can this be quantified? Does the distribution between the different sources vary during the day and during the season, and can this be quantified?**

Collected water can originate from various sources: (i) surface water, such as ditches, ponds, small streams, (ii) puddles, (iii) guttation water, (iv) dew drops or (v) honey dew. All sources may occur in or outside the treated field. Surface water generally is a permanent source, while puddles generally evaporate and seep into the soil after several hours or days. Guttation water and dew drops generally dry up after a couple of morning hours, while honey dew may persist during the day.

The WG intends to further detail the various sources described above and to try to quantify these as much as possible by performing an extensive literature search and on the basis of submitted dossier studies.

### **Preliminary estimate of the water consumption of adult bees**

This data for estimating the water consumption in an adult comes from Free & Spencer Booth (1958). Although this is the same data used to calculate the values for adult water consumption in the 2013 document, the interpretation is different. The main change is a reduction in the estimation of the amount of water an adult bee consumes from a source of pure water, independent of water from any other source. The estimate in the 2013 document was 11.4 µl per day, per bee, this has been revised to 0.7 µl per day, per bee (see supplementary information below for more detail).

The difference between the two estimations comes from (i) acknowledging that the experiment which generated this data was not designed to determine water consumption and contained some methodological issues which were not considered in the 2013 document and (ii) acknowledging that a large amount of a bees water requirements are met by consuming a liquid diet of honey/nectar.

## **Q1.2. What different sources cover the water needs of larvae? Can this be quantified? Does the distribution between the different sources vary during the day and during the season, and can this be quantified?**

Consideration on how the larvae feed:

- The eggs hatch into larvae that are destined to become workers, queens or drones.
- Larvae are provided with food by the worker bees (nurses).
- Larvae are fed either Worker or Royal Jelly which is a product of the hypopharyngeal and mandibular glands (Winston, 1987).
- The composition of Worker and Royal Jelly is similar for proteins, sugar and lipids within the first three days (Von Rhein, 1933; Brouwers et al., 1987). The water content is uniform and >60% (the constancy of moisture content is assured, in the hive, by the continuous provision of fresh supplies of this substance by nurses, by the natural hygroscopicity of the Royal Jelly and the entire efforts to maintain a level of ambient moisture (Sabatini et al., 2009).
- Worker larvae are fed solely from pharyngeal secretions for the first three days, and on the 4<sup>th</sup> and 5<sup>th</sup> day some honey/nectar and beebread are introduced to the diet

(Haydak, 1943; Shuel and Dixon, 1959; Haydak, 1968, 1970; Kunert and Crailsheim, 1988; Malone and Pham-Delegue, 2002).

- Queen larvae are fed from pharyngeal secretions for their entire larval development (Shuel and Dixon, 1959). Worker larvae probably have the highest risk of exposure to external contamination (pollen, unprocessed nectar, water as their diet it supplemented with bee bread and honey after the third day).

The claim that honeybees use water to dilute the larval food is made quite frequently (Seeley, 1995) and is based on the difference in water content between honey (~18%) and larval food (up to 70%). However, larval food, at least within the first three days, consists solely of Worker or Royal Jelly, and reference to the use of water to dilute the Royal Jelly has not yet been found.

Up to now the WG assumes the water need of larvae is covered exclusively by the Worker or Royal Jelly in first 3 days and the next 2 days there could be water added to dilute honey (depending on the sugar concentration).

The WG will consider to revise the estimation of the water consumption by larval worker honey bee reported in the EFSA, 2013, by taking into account the food consumption, the composition of the Royal Jelly given to the workers as described in Wong et al 2016 and the available data on the pesticide residue in Royal Jelly. As this is solely made up of undiluted glandular secretions, and there are only trace levels of contamination in royal jelly (see section 4.2.3), the WG will assume that the presence of contaminated water in the first three days may be set to zero.

**Q1.4. How many days of the year adult honey bees or larvae consume water collected by the water foraging bees (i.e. not only from the water contained in nectar or honey)? Which conditions (both environmental and bee behavioural) determine whether water is collected?**

In order to decide how relevant the so-called water scenarios in the EFSA, 2013 are the WG intends to attempt to estimate the number of days during the year that bees collect water for consumption (i.e. not cooling), so to determine the temporal probability of occurrence in time that at least part of the pesticide intake by bees or larvae originates from collected water. If water collection would occur in less than 10% of the days in the year (excl. overwintering), one might argue that the water scenarios are not relevant. Water collection is governed by a number of conditions, such as environmental conditions as well as bee behaviour. Also bee-keeping practices may intervene. E.g. in the text under question 1.1 we already mentioned the conditions of (i) nectar scarcity, (ii) low temperature, below 9°C, or (iii) rainfall, that prevent foraging bees to collect nectar. While conditions (ii) and (iii) relate to environmental conditions, condition (i) is influenced by bee-keeping practices: Beekeepers will always try to place their hives in locations where nectar is available, thus to avoid nectar scarcity.

The WG will try to illuminate the conditions that drive bees to collect water for consumption in the hive by performing an extensive literature search. In particular, the WG intends to describe the conditions required for water collection by bees in order to attempt to estimate the number of days they collect water for consumption.

The proposed approach to answer the question 2 is outlined below.

**Q2 Is the exposure to pesticides by collected water (esp. guttation water) a relevant scenario?**

## Q2.1 Adult honey bees: what is the proportion of the daily pesticide intake via consumption of collected water compared to the daily pesticide intake via nectar and pollen

The oral exposure, i.e. pesticide intake from collected water is defined below, for the adult bee (acute and chronic toxicity) as well as the brood (chronic toxicity only);

$$Cons_{pesticide} = W PEC \quad \text{Eq 1}$$

With

$Cons_{pesticide}$  = Pesticide Consumption per individual (mass in  $\mu\text{g}$  per bee or larvae);  
 $W$  = Water consumption per individual (volume in  $\mu\text{L}$  per bee or larvae);  
 $PEC$  = Concentration of pesticide in the consumed water (mass of pesticide divided by volume of water, e.g.  $\mu\text{g}/\mu\text{L}$  or  $\text{mg}/\text{L}$ ),

Note that  $W$  and  $PEC$  are daily values for the assessment of the acute toxicity for adults and time-weighted average values over 10 and 5 days for the assessment of the chronic toxicity for adults, resp. larvae. Definition of the Exposure Assessment Goals for the pesticide intake indicated that daily values or time weighted average values over 5 or 10 days period need to be considered. So, the concentration in the consumed water is determined as the average concentration in the water entering the hive for consumption. The daily PEC value may consist of a weighted average value of e.g. guttation water and puddle water, accounting for a worst-case situation when guttation water is used when present (morning hours).

The WG assumes that the consumption of the collected water may occur in some circumstances (objective of the Q1). Where those circumstances occur, the WG deems not defensible to consider only the uptake of pesticide via water. In such cases the total oral intake of pesticides should take into account the other sources (i.e. residue intake via consumption of pollen and nectar at the same time as residue intake via consumption of collected water). This can only be assessed by considering the correct total intake,  $Cons_{pe}$  via:

$$\begin{aligned} Cons_{pe} &= W PEC + Cons_{pollen} Conc_{pollen} + Cons_{nectar} Conc_{nectar} \\ &= AR (W RUD_{water} + Cons_{pollen} RUD_{pollen} + Cons_{nectar} RUD_{nectar}) \quad \text{Eq 2} \end{aligned}$$

with

$AR$  = application rate (mass of pesticide divided by area, e.g.  $\text{kg}/\text{ha}$ )  
 $RUD_{water}$  = concentration in water (mass of pesticide divided by volume of water, e.g.  $\text{mg}/\text{L}$ ) at an application rate of  $1 \text{ kg}/\text{ha}$   
 $Cons_{pollen}$  = consumption of pollen per individual (mass in  $\mu\text{g}$  per bee or larvae);  
 $Cons_{nectar}$  = consumption of nectar per individual (mass in  $\mu\text{g}$  per bee or larvae);  
 $RUD_{pollen}$  = concentration in pollen (mass of pesticide divided by mass of pollen, e.g.  $\text{mg}/\text{kg}$ ) at an application rate of  $1 \text{ kg}/\text{ha}$

$RUD_{nectar}$  = concentration in nectar (mass of pesticide divided by mass of nectar, e.g. mg/kg) at an application rate of 1 kg/ha

### Calculation of $Cons_{pesticide}$ .

The WG considers to first make indicative calculations in order to demonstrate (i) relative importance pesticide from collected water versus from total amount of collected nectar, (ii) relative importance pesticide from collected water versus from pollen (nurse/forager/brood), (iii) whether also nectar and pollen consumption need to be maintained in ranking total residue intake during water consumption or only water consumption (i.e. Eq 1 or Eq 2 above), (iv) effect of guttation only during morning hours on daily PEC, and (v) to underpin which distribution is most important for RA (consumption or PEC) ?

Later on, Monte Carlo simulations could possibly be done, with probably a single value for water consumption  $W$ , and stochastic distributions for concentration in water  $PEC$  (combine 9-12 am guttation concentration distribution + 12-16 pm other, e.g. puddle concentration into 1 daily distribution, probable worst case situation), plus distributions in maybe pollen consumption,  $Cons_{pollen}$  and concentration in pollen,  $Conc_{pollen}$  as well as in nectar consumption,  $Cons_{nectar}$  and concentration in nectar,  $Conc_{nectar}$ .

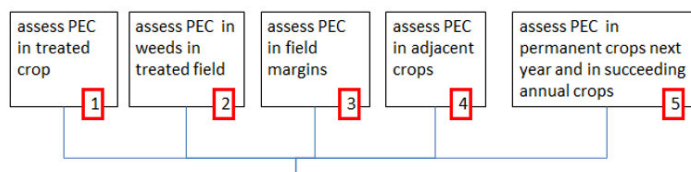
Extensive literature search will be done e.g. the field of occurrence and concentrations in guttation water in literature and dossier studies (e.g. Christl et al, 2019). Calculations with the repaired FOCUS surface water scenarios will give estimations for concentrations in puddle water and small surface water bodies.

### Scenarios and application techniques

In principle 5 water scenarios are possible and 3 types of applications (Fig 1, EFSA, 2013). These are (i) the treated crop, (ii) the weeds in the treated field, (iii), the plants in the field margin, (iv) the adjacent crop and (v) the permanent crops in the next year or in succeeding annual crops. The 3 types of application techniques are (i) seed treatment, (ii) spray (downward, or side- and upward) and (iii) granules.

## Water scenarios and application types

- In principle 5 scenarios possible
- times 3 application types: seed treatment, spray (downward, or side- and upward) and granules



**Figure 1. Water scenarios and application types.**

Note that the application technique will influence the concentration in the collected water, e.g. for seed treatment concentration in guttation water is expected to be significant, but puddles and surface water are expected to be not or hardly contaminated. For spray applications both guttation water and puddles and surface water may contain pesticides.

The indicative calculations mentioned above will focus on the water scenario where the origin of the water is from within the treated crop and seed treatments (expected to be worst case with respect to guttation).

## **Q2.2 Larvae: what is the proportion of the daily pesticide intake via consumption of collected water compared to the daily pesticide intake via (royal or worker) jelly, nectar and pollen?**

A similar approach could be followed for larvae. Note that exposure via royal jelly has been treated separately in the protocol (see section 4.2.3).

## **Q2.3. Can the occurrence of guttation be characterised over the day ? This is expected to be a function of (i) crop type, (ii) BBCH crop development stage, (iii) , and environmental conditions.**

As the target PEC is a daily value, and guttation does not occur during the entire day, the PEC will never consist of the  $PEC_{\text{guttation}}$  only, but a weighted average value over the entire day is used. Therefore, it is important to have estimations of the duration of guttation as a function of crop and BBCH stage and, if possible also as a function of environmental conditions. E.g. on damp, cold days, guttation is expected to last longer than on sunny, dry mornings. To obtain this information, the WG will perform an extensive literature search and will use dossier studies.

## **Q2.4. Can the concentration of pesticides in the guttation water be quantified as a function of time after application?**

**This is expected to be a function of (i) compound, (ii) crop and BBCH crop development stage, (iii) application technique (seed treatment or spray-downward or up- and sideward) and (iv) application timing**

For the indicative calculations mentioned above measured concentrations in guttation water are available (e.g. Christl et al, 2019). However, a distribution in concentration values in guttation water would be useful to estimate the pesticide uptake by guttation water. On one hand, experimental data are available and will be retrieved, on the other hand it will be considered whether regulatory modelling results (e.g. soil pore water concentration in EU FOCUS soil exposure scenarios) could be used.

## **Q2.5 How do the pesticide concentrations in guttation water, puddle water and surface water compare in general?**

Concentrations in puddles and surface water can be obtained by following the regulatory procedure to estimate exposure in the EU FOCUS surface water scenarios. Concentration in puddles filled by runoff are approximately maximally 0.5 mg/L (compound  $K_{om} = 30$  L/kg,  $DegT_{50,soil} = 10$  d, 1 kg/ha, see e.g. FOCUS Repair, Fig 1). Concentrations in FOCUS ditches, streams or ponds are lower than concentration in puddles, as their water is a mixture of runoff of treated and untreated fields, and thus the concentration in runoff water will be diluted. The concentration in over sprayed puddles is approximately 1 mg/L (1 kg/ha, 10 cm water depth). Concentrations in guttation water may be significantly higher, but a careful and systematic comparison needs to be done, taking into account that the application rates that are often expressed as mg/seed need to be converted to be comparable (e.g. both in kg/ha).

### **Supplementary Information to the preliminary estimation of daily adult honey bees water consumption**

The rationale for making the revised water consumption estimates is detailed below and based on the data from Free and Spencer-Booth (1958):

The WG first tried to exclude any temperature treatments or groups that would be unsuitable for determining water consumption. The original paper suggested that the death rates were at their lowest and most stable between 25-35°C (Supplementary Figure 2A) but that the two smallest groups showed anomalous spikes in mortality at 30°C (data not shown). If we take that as correct, then we should assume that the temperatures in this range were the least likely to put the caged bees under thermal stress; so, we should exclude groups <25° and >35°C for estimating water consumption. Given the unexplained variation in the death rates in the two smallest groups these have also been excluded.

At lower temperatures the bees require carbohydrates to generate heat which likely explains the dramatic reduction in sugar intake in the warmer cages. The dashed line in Supplementary Figure 2B indicates the lower boundary for the daily sugar intake for a forager bee (lower boundary for a nurse bee is very similar at 34 µg/day) recorded in EFSA, 2013 which shows just how different this experiment is from bees in a hive. However, we might still be able to gain some insight into the water requirements for bees. The rate of decline in sugar intake at higher temperatures appears to drop off between 25-30°C, i.e. sugar consumption drops by 20 µg between 15 and 25°C but only drops by 5 µg between 25-35°C, indicating that these values (somewhere between 5-15 µg per day) likely reflects the amount of sugar required to keep a bee alive, when we don't impose any energetic costs that require supplementary feeding e.g. in hive tasks or having to generate metabolic heat. This agrees with the mortality data.

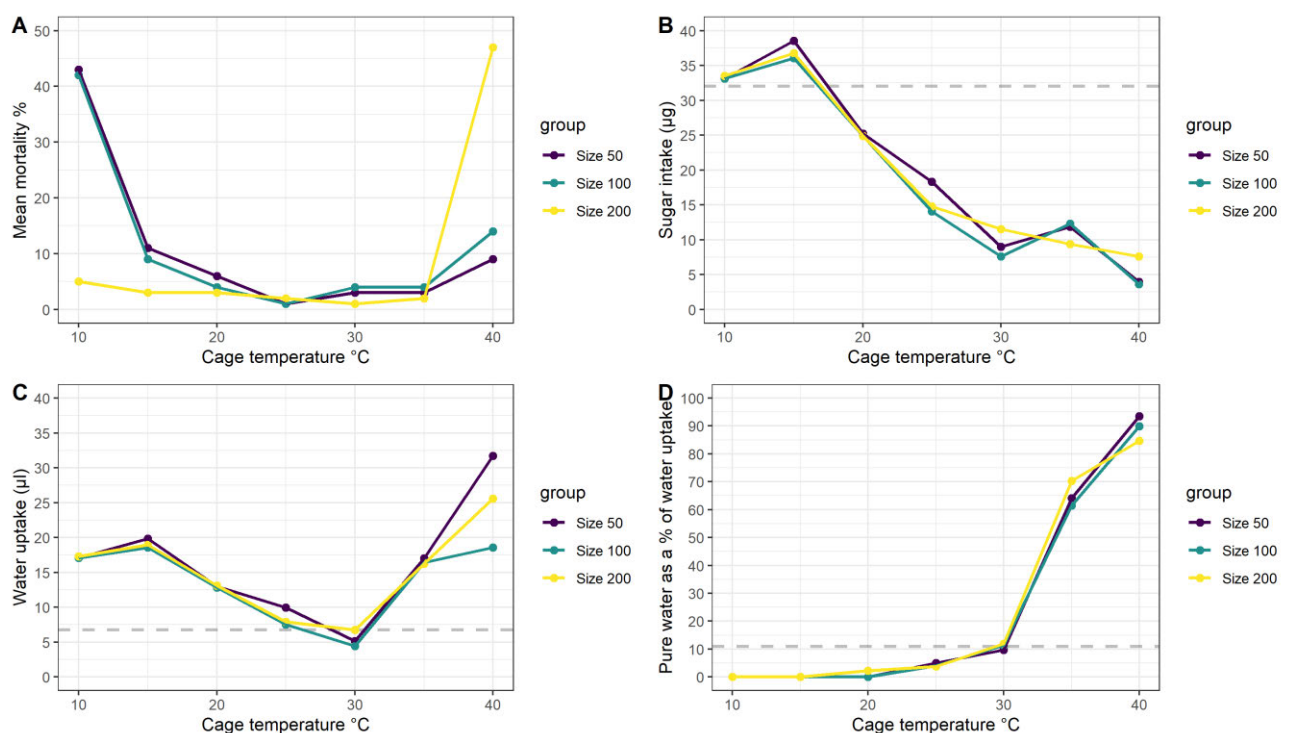
Supplementary Figure 2C shows the average total water uptake per bee, which is the sum of the water in the 66% sugar syrup, and the pure water provided to the caged bees. Supplementary Figure 2D shows the uptake of pure water relative to the water from the sugar syrup. At low temperatures (<25°C), the water intake is relatively high because the bees need to increase their sugar intake by consuming large volumes of syrup and ignoring the foraged water. When bees take on more water than they need, they simply excrete the excess. The steady decline in sucrose and water uptake at temperatures up to 30°C show that the bees reducing their food intake when they don't require the nutrients from the food. The water intake was minimised at 30°C indicating that this is likely as good an estimation of a bees the water requirements as we can get from this experiment.

There is an argument that we should use the values from 35°C treatment, as these temperatures are commonly found within the hive, however, this would be unwise. The



experimental conditions in this study were highly unrepresentative of the conditions in the hive or whilst foraging; the bees were held in a temperature-controlled cage with no in-hive tasks or foraging tasks and the cages had no humidity control – both of these factors are highly likely to affect a bees energy requirements and transpiration rates which in turn impact on a bees water requirements. We know that the hive is maintained in a state of homeostasis, where both temperature and humidity are regulated. If we assume that honeybees have evolved so they are not constantly in a state of stress (such as might be indicated by a tripling in the amount of water uptake between the 30°C and 35°C treatments) then we need to use measurements where there is minimal evidence of either cold stress (causing increases in syrup consumption) or heat stress (causing rapid increases in water consumption).

The WG is choosing to use the data generated from the 30°C treatment which indicates that a bee requires between 5.124-6.750 µl water per day. From this total water requirement, the bees consume between 10-12% come from pure water leading to the revised estimate of 0.7 µl per day, per bee.



**Figure 2: Supplementary information. Mortality rates for each temperature treatment (Plot A), the average daily amount of sucrose consumed per bee with the dashed lines indicating sugar requirements of a forager bee in a hive (Plot B), average daily water uptake per bee with my estimate for the amount of water a bee requires to survive (Plot C), the proportion of pure water as a percentage of total water uptake and my estimated value for the proportion of water that comes from pure water (Plot D).**

### Miscellaneous:

The results are reported in mm<sup>3</sup> so need to be converted to a volume and mass for water and sugar. Here, 1 mm<sup>3</sup> is equal to 1 µl of water and 1 mm<sup>3</sup> is equal to 1 µg of sugar.

The description of the sugar syrup in the paper was 2-parts sugar to 1-part water reported as a w/w% - so a 66% sugar water solution.

There were five group sizes (10, 25, 50, 100, 200 individuals) the smaller group sizes showed highly variable responses and high stochastic mortality. These were removed.

The observations could not distinguish between water that the bees drunk (e.g. consumed and then absorbed through the gut) from water that was used for other purposes e.g. cooling. The WG refers to the use of water as water uptake.

## 4.2. Hazard assessment

### 4.2.1. Inter-species toxicity endpoint difference and related assessment factors

#### 4.2.1.1. Background of the issue

In the current EFSA Guidance Document (2013) the risk assessment is carried out separately for honey bees, bumble bees, and solitary bees.

**Honey bees:** In Europe, honey bees are represented by a unique species (*Apis mellifera*). Standard toxicity tests with this species are always required for pesticide risk assessment. Hence, the available data can be directly used for estimating the risk. Despite different subspecies of *A. mellifera* exist, it is assumed that the difference in sensitivity between subspecies is negligible and no additional uncertainty factor are necessary.

**Bumble bees:** there are 68 species of bumble bees in Europe. Standard acute test methodologies are available for *Bombus terrestris*, however these studies are not mandatory for pesticide risk assessment. When data with *B. terrestris* are not available, the EFSA GD considers that the toxicity data on honey bees (surrogate species) may still be used for addressing the risk to bumble bees, provided that an additional Assessment Factor (AF) of 10 is used in the risk assessment i.e. the endpoint for *Bombus terrestris* is conservatively assumed 10 time lower than the one for *Apis mellifera*. This AF is derived from Arena and Sgolastra (2014). Even in cases where data on *Bombus terrestris* are available, the EFSA GD suggests using in the risk assessment an additional AF of 5 which should cover, among other uncertainties, also the inter-species differences among bumble bees; i.e. extrapolation from *Bombus terrestris* to any other species of bumble bees.

**Solitary bees:** there are almost 1900 solitary bee species in Europe, comprising taxa that are very diverse. Standard test methodologies for assessing the toxicity of chemicals are not available, despite some attempts have been made with at least a few species (*Osmia spp.*, *Nomia melanderi*, *Megachile rotundata*). When toxicity data with solitary bees are not available (almost all cases), the EFSA GD considers that the toxicity data on honey bees (surrogate species) may still be used for addressing the risk to solitary bees, provided that an additional Assessment Factor (AF) of 10 is used in the risk assessment i.e. the endpoint for the 'standard solitary bee species' is conservatively considered 10 time lower than the one for *Apis mellifera*. This AF is derived from Arena and Sgolastra (2014). Even in cases where data on one species of solitary bees are available, the EFSA GD suggests using in the risk assessment an additional AF of 5 which should cover the inter-species differences among solitary bees; i.e. extrapolation from the tested species to any other species of solitary bees.

The assessment factor currently used can be summarised in (table 7).

**Table 7. Assessment factor in the EFSA, 2013**

Bee group	AF-1: covers endpoints' difference between honey bee and standard species of the group	AF-2: covers endpoints' difference between standard species of the group and all other species within the group
Honey bee	N/A	1 (one species only)
Bumble bee	10	5*
Solitary bee	10	5**

\* note that this factor is also supposed to cover for other uncertainties

\*\* There are currently no standard test species for solitary bees, however assuming that the endpoint for one species would be available, this factor should still be applied in order to cover for difference in sensitivity with other solitary bee species.

An update of this assessment factors was considered by the WG. To this end, information is needed to cover two sources of known uncertainty, which can be summarised by the following questions:

**Q1: What is the ecotoxicity endpoint difference between *Apis mellifera* and the standard species of the group (for both bumble bees and solitary bees)?**

**Q2: What is the ecotoxicity endpoint difference between standard species of the group (for both bumble bees and solitary bees) and the remaining species of the group?**

Both questions relate to ecotoxicity endpoints derived from tests with pesticides.

#### 4.2.1.2. Proposed approach

All these AFs could be in principle be refined with suitable data. The scope of this activity would be which factors could indeed be refined and to what extent.

While the comparison of sensitivities should account for differences in body weight, this parameter is not measured in the vast majority of cases, as historically the toxicity endpoints are expressed in mass substances/bee.

In lack of study-specific measurements, one could only rely on default (fixed) body weight values. However, this would not provide any benefit for the implementation of the AFs, as the inverse of the 'conversion factor' (body weight ratio) used for calibrating the AF would need to be applied when estimating the missing endpoint. The WG noted however that in case study-specific body weight data will be routinely available in future, a proper sensitivity factor considering the body weight differences could be worked out.

Data on toxicity tests with bees are likely to be found both in pesticide dossier and in literature studies. The relative usefulness of the two sources for the different questions is discussed below.

The limiting factor in terms of data availability is never honey bees. As such, the initial focus will be on substances for which data on other bee species are available. Retrieval of honey bee data for any pesticide is rather straightforward and hence it is not considered further. However, once identified the pool of substances for which a comparison can be performed, endpoints will need to be retrieved for honey bees as well: in this case, the priority will be given to dossier studies. The study appraisal and the data extraction (see below) will be the same for all bee groups.

### *Objective 1: Difference between honey bee to standard species of another bee group*

In principle, objective 1 could be addressed by considering, for a suitable number of substances, the endpoint ratio between *Apis mellifera* and other standard species for bumble bees and solitary bees. If enough data are available, a certain percentile (e.g. 50<sup>th</sup>, 90<sup>th</sup>, 95<sup>th</sup>, absolute worst-case, etc.) of the ratios' distribution could be selected as AF. For bumble bees the selection of the standard species (i.e. *Bombus terrestris*) is straightforward. On the contrary, for solitary bees this is more complex, due to the lack of recognised standard test guidelines. Hence, the selection of the 'standard species' for solitary bees will be made only after considering the relative abundance of data for the different species.

### *Objective 2: difference from standard species to all other species of the same group*

For objective 2, toxicity data on multiple species should be available for a certain number of substances. If those data were available, it would be possible to calculate, for every substance, the ratio between the standard species endpoint and another value (dose) protective for all the tested species (e.g. most sensitive species, HD<sub>5</sub> derived from an SSD, etc.). Then, an appropriate percentile of this ratio distribution could be selected as an AF to be implemented in the risk assessment. As commented in the section above, for bumble bees this procedure would be, at least theoretically, quite straightforward. Nevertheless, for solitary it wouldn't be as clear, due to the lack of a recognised standard test species.

The data retrieval, as well as study screening, appraisal, and data extraction will fully overlap for the two objectives. The only difference defined a priori would be that, for objective 1 and bumble bees, the focus will be on *Bombus terrestris* only.

## Source of the data

Useful data are available in pesticide dossiers. A systematic check on 241 substances has been already performed, resulting in bumble bee data for 25 substances, and solitary bee data for 2 substances.

Some relevant data are also expected to be found in the open literature. Recently, a quasi-systematic review on the topic, covering material published until June 2019, has been performed by Lewis and Tzilivakis (2019). This will be used as a basis for the comparison.

In order to have a more systematic check of what is available, the WG decided to rely on the ECOTOX database maintained by the US EPA. This database aims at collecting endpoints from published literature. It currently contains almost 1 million records. Details of the suggested query to be run in the database are reported below. As the database is updated at discrete time-intervals, the time between the last update and the collection of data should be covered by a targeted literature search: this time is usually less than six months. The details of the query used for the ECOTOX database (table 8) will also be used for designing a search string for the integrative literature search.

**Table 8. Tentative query input for the US EPA ECOTOX database**

Query parameter	Selected values
Habitat	Terrestrial
Chemicals	(empty)
Effect Measurements	Mortality Group
Endpoints	Concentration Based Endpoints EC/ED xx (all % values) LD50 LC50 LC/LD xx (all % values)

	EC50 ED50 NOEC NOEL
Species (based on a preliminary check of relevant taxa currently included in the database)	Andrena erythronii Andrena sp. Andrenidae Apidae Augochlorella sp. Augochlorella striata Bombus auricomus Bombus impatiens Bombus lapidarius Bombus lucorum Bombus occidentalis Bombus sp. Bombus terrestris Bombus terrestris ssp. audax Bombus terricola Bombus terricola ssp. occidentalis Bombus vosnesenskii Frieseomelitta nigra Frieseomelitta sp. Halictidae Halictus ligatus Halictus rubicundus Halictus sp. Hoplitis adunca Hoplitis sp. Hymenoptera Hypotrigona ruspilii Lasioglossum sp. Lasioglossum zephyrum Megachile rotundata Megachile sp. Megachilidae Melipona beecheii Melipona quadrifasciata Melipona quadrifasciata ssp. anthidioides Melipona scutellaris Melipona sp. orned Bee - Melissodes bimaculata Melissodes sp. Nannotrigona perilampoides Nannotrigona sp. Nomia melanderi Nomia sp. Osmia cornifrons Osmia cornuta Osmia lignaria Osmia rufa Osmia sp.

	Partamona helleri Partamona sp. Peponapis pruinosa Peponapis sp. Plebeia droryana Plebeia sp. Scaptotrigona postica Scaptotrigona sp. Tetragonula iridipennis Tetragonula sp. Trigona sp. Trigona spinipes
Test Conditions	Test Locations Lab
Publication Options	(empty)

### Eligibility criteria

The screening of the studies from the literature search should only focus on retaining laboratory experiments where (non-*Apis*) bees are individually dosed with chemicals/pesticides for the derivation of a standard mortality endpoint (LD50, LDD50, NOED, etc.). Data on acute (oral and contact), chronic, and larvae will be retained.

Only definite endpoints (not unbounded values) will be considered, as comparison involving unbounded values cannot provide an accurate quantification for the differences.

### Study appraisal

Irrespective of the origin of the data, the studies should be appraised for their internal validity and precision using criteria in line with the relevant OECD guidelines. Considering the focus on laboratory toxicity data, an evaluation of external validity is not needed.

Criteria to be considered for internal validity

- KEY QUESTION: Is the mortality in the control <10% (acute tests) or 15% (other tests)?
- If a solvent was used, is a solvent control present?
- Were the test organisms properly acclimatised to the study setup before the exposure started?
- Is the health status of the organisms appropriate (are they disease-free and not being stressed before)?
- Is the sensitivity of the system tested? (e.g. positive control)<sup>4</sup>
- Is the duration of the observation appropriate? (e.g. 2-4 days for acute, 10 days for chronic, etc.)
- Is the exposure duration appropriate? (e.g. single exposure for acute, 10 days for chronic, etc.)
- Was the purity of the active substance or the active substance content in the formulation accounted for in the endpoint derivation?
- Is exposure characterised by analytical measurements (residues or confirmed dose – chronic tests only)?

<sup>4</sup> This is only applicable if the toxicity of a reference substance to the tested organism is known.

- Is the evaporation of the sucrose solution checked and accounted for (chronic tests with adult bees only)?

#### Criteria to be considered for precision

- Is the test replication appropriate? (e.g. 30 bees per tested concentration for adult bees)
- Is the number of tested concentration/doses appropriate? (at least 5)
- Is a correct spacing adopted between tested doses/concentrations? (geometric series with a factor not exceeding 2-3)
- Is the endpoint derivation suitable (e.g. fit of dose-response model)<sup>5</sup>

#### Data extraction

Only mortality data will be extracted (lack of emergence for larvae is considered equivalent to mortality). A tentative model for the data extraction is reported below.

**Table 9. Tentative data model for the data extraction**

Parameter	Data type
Study ID	Key study identifier
Author	Free text
Journal/Volume/Issue	Free text
Guideline (if available)	Picklist <ul style="list-style-type: none"> <li>• OECD TG 213</li> <li>• OECD TG 214</li> <li>• OECD TG 246</li> <li>• OECD TG 247</li> <li>• OECD Guidance Document 239</li> <li>• None</li> <li>• Other (free text)</li> </ul>
Species	Picklist (to be implemented after preliminary analysis of availability)
Bee group	Picklist <ul style="list-style-type: none"> <li>• Honey bees</li> <li>• Bumble bees</li> <li>• Solitary bees</li> </ul>
Life stage	Picklist <ul style="list-style-type: none"> <li>• Adult</li> <li>• Larva</li> </ul>
Tested substance	Picklist (to be implemented after preliminary analysis of availability)
GLP	TRUE/FALSE
Endpoint time scale	Picklist <ul style="list-style-type: none"> <li>• Acute</li> <li>• Chronic</li> <li>• Single dose (larvae)</li> <li>• Repeated dose (larvae)</li> </ul>

<sup>5</sup> This criterion is not relevant if raw data are available, as in such case the endpoint could be re-estimated.

	<ul style="list-style-type: none"> <li>• 5-days</li> <li>• Other</li> </ul>
Test duration [days]	Float
Exposure type	Picklist <ul style="list-style-type: none"> <li>• Contact (direct application)</li> <li>• Contact with treated surface</li> <li>• Oral</li> <li>• Oversprayed</li> <li>• Other (free text)</li> </ul>
Number of tested doses	Integer
Bees per treatment dose	Integer
Bees in the control(s)	Integer
Effect	Picklist <ul style="list-style-type: none"> <li>• Mortality</li> <li>• Emergence failure</li> <li>• Development failure</li> </ul>
Endpoint type	Picklist <ul style="list-style-type: none"> <li>• LD50</li> <li>• LDD50</li> <li>• NOED</li> <li>• LC50</li> <li>• NOEC</li> </ul>
Endpoint qualifier	Picklist <ul style="list-style-type: none"> <li>• N/A</li> <li>• &lt;</li> <li>• &gt;</li> <li>• ≥</li> <li>• ≤</li> </ul>
Endpoint value (mean)	Float
Endpoint lower CL (95%)	Float
Endpoint upper CL (95%)	Float
Endpoint unit	<ul style="list-style-type: none"> <li>• µg/bee</li> <li>• µg/bee/day</li> <li>• µg/bee/dev. period</li> <li>• µg/L</li> <li>• µg/kg</li> <li>• µg/cm<sup>2</sup></li> </ul>

#### 4.2.2. Extrapolation factor beyond the tested rates for less toxic substances

##### 4.2.2.1. Background of the issue

For five of the six currently available laboratory OECD tests on bees (TG 213, TG 214, TG 246, TG247, and Guidance Document 239) the main endpoint selected in the previous EFSA Guidance Document (EFSA, 2013) is either an LD<sub>50</sub> or an LDD<sub>50</sub>. In all the aforementioned guidelines, when a low toxicity is expected, there is the possibility to carry out 'limit dose' tests, i.e. bioassays performed with only one dose. Such limit dose for the acute tests is consistently reported to be 100 µg a.s./bee. Hence, it is often inferred that, even in complete dose-response studies, the highest dose should not be higher than 100 µg a.s./bee.

It follows that, for less toxic substances, the endpoint is often represented by censored (unbounded) values (e.g. LD<sub>50</sub> > 100 µg a.s./bee). When this is the case, it is common



practice in ecotoxicology to consider the lower bound of the censored value as input in the risk assessment (e.g.  $> 100 \mu\text{g a.s./bee}$  is considered equal to e.g.  $100 \mu\text{g a.s./bee}$ ), as this represents a worst-case. As such, uses with rather high application rates are often triggering a high risk at the lower tiers, despite zero or very low effects were recorded in the toxicity study. In order to ensure a fair risk assessment, and not to penalise substances with low toxicity, a more realistic description of the hazard is needed in these cases.

The WG considered working out an extrapolation factor pending on the actual effect observed at the highest tested dose (in case this is  $< 50\%$ ). In other words, the present task should answer the following question.

**What is the appropriate extrapolation factor to apply to unbounded endpoint pending on the effect observed at the highest tested dose?**

#### 4.2.2.2. Proposed approach

The present issue was already considered for the risk assessment of birds in the EFSA Scientific Opinion (EFSA PPR Panel, 2008). In the Opinion, an approach has been proposed and detailed in the Appendix 5. Later this has been adopted in the following Guidance Document (EFSA PPR Panel, 2009) and has been used in pesticide risk assessment since.

This approach considers the possibility of working out extrapolation factors for situations where there is either zero or single (only one dead animal) mortality  $x$  at the limit dose  $d$ . By assuming a generic probit dose-response curve, the estimation of the extrapolation factors depends on:

- The  $n$  number of animals tested at the limit dose.
- The required confidence level  $c$ .
- The slope  $b$  of a generic probit dose-response curve fitted to the log of the dose.

While the number of animals tested depends on the test design and the required confidence level should simply be selected (for birds, this was fixed at  $c=0.5$ ). In order to get a generic slope  $b$  of the probit curve, real dose-response relationships were collected.

The WG considered another, very similar approach for estimating the extrapolation factors, which is based on the theory of the log-logistic dose-response models. These models are generically described by four parameters (although asymmetric models with a fifth parameter exist) explaining the lower limit, the upper limit, the slope, and the potency. Dose-response curves for corrected mortality data are always bound between a lower limit of 0 (i.e. the mortality observed in the control) and an upper limit of 1 (total mortality). Hence, the relation between any  $\text{LD}_x$  and the  $\text{LD}_{50}$  is uniquely affected by the slope parameter. The lower the slope, the bigger is the ratio  $\text{LD}_{50}/\text{LD}_x$  (assuming  $x < 50$ ). Hence, for any given slope, one could work out extrapolation factors for all possible  $\text{LD}_x$ .

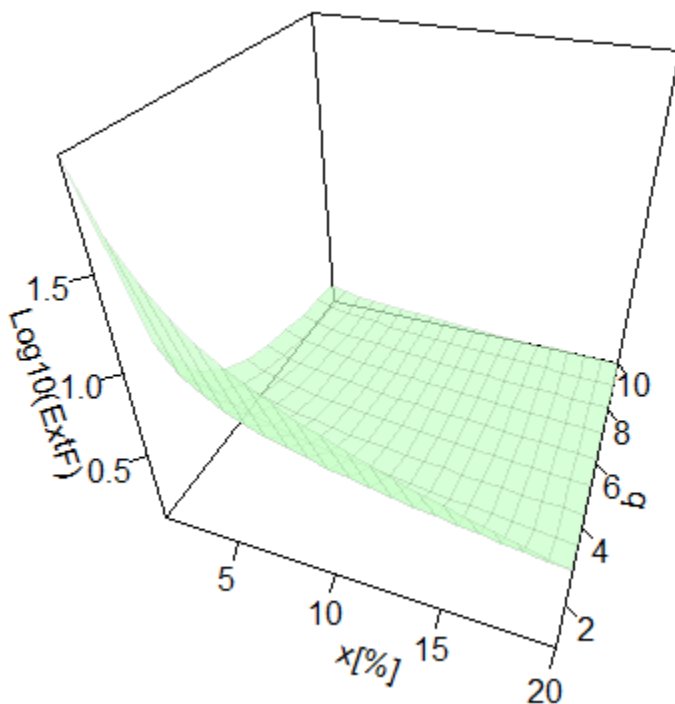
As the previous method, also the present needs to rely on the concept of a generic slope. However, provided that a suitable number of studies (and related dose-response) are available, one could analyse the distribution of the slopes and pick a suitable generic one (e.g. mean, 90<sup>th</sup> percentile, etc.).

Then, one could assume that the observed corrected mortality at the limit dose is the best estimate of the true mortality due to the substance's toxicity (e.g. if there is 5% corrected mortality at the limit dose, this is immediately considered as the best  $\text{LD}_5$  estimate). By fixing the slope  $b$ , one fixes also the  $\text{LD}_{50}/\text{LD}_x$  ratio, which corresponds to the extrapolation factor

to be applied to the limit dose at which  $x\%$  corrected mortality is observed. Expressed as a mathematical formula, this is:

$$ExtF = d * \frac{LD_{50,b}}{LD_{x,b}}$$

Where  $x$  represents a percentage mortality. The relation between slope [1-10], the level of mortality  $x(\%)$  [1-20%] at the limit dose  $d$ , and the Log10 of the extrapolation factor is illustrated in the picture below.



**Figure 3. The relation between slope ( $b$ ) [1-10], the level of mortality ( $x$  [%]) [1-20%] at the limit dose  $d$ , and the Log10 of the extrapolation factor ( $ExtF$ ) estimated on the basis of the log-logistic model.**

Both methods considered rely on the concept of monotonic dose-response. In cases where monotonicity is not fully clear (e.g. higher level of mortality observed at doses lower than the limit one), the applicability of extrapolation factors should be carefully considered.

While in principle the method used for birds has the advantage to be able to consider the level of confidence, it has also the big disadvantage that analytical solutions are only available for cases when only 0 or 1 animal are dead at the limit dose. For instance, with 2 bees dead out of 30, the mortality level would still be 6.7%, but the limit dose would in this case be used as surrogate of the LD50 without any extrapolation factor.

Hence, the WG expressed their preference for the methodology based on the log-logistic dose-response models. The WG also expressed the preference to discretise the extrapolation factor for intervals of effects seen at the limit dose, i.e. fixed extrapolation factors for effects 0-10%; 10-20%; 20-30%; 30-40%.

#### 4.2.2.3. Slope data collection

The only data collection needed for addressing this issue regards slopes of dose-response relationships. Details on how this operation will be conducted are reported below. This collection will consider both acute and chronic data. It was also to differentiate between herbicides, fungicides and insecticides (however most of the suitable data are expected to be from insecticides, see eligibility criteria below). If feasible, information on bumble bees (mainly *Bombus terrestris*) and solitary bees will be collected as well. However, as dose-response curves will be only available for a limited number of substances, this collection will chiefly be used to cross-check the possibility for extrapolation from honey bees to other bee groups.

#### Source of data

The main source of data will be pesticide dossier studies, as effects at the individual dose are always available in the studies submitted for regulatory purposes. As complementary information, the studies collected on non-*Apis* bees (see section 4.2.1) will also be checked.

#### Eligibility criteria

Studies should respect the following basic criteria

- 1) Be valid according to the relevant OECD standard
- 2) Report effect at the different tested doses
- 3) Presenting a range of effects that allow a reliable estimation of the dose-response (monotonic effects from <10 to >50%).

Criterion 3 will likely bias the relative contribution of insecticides, fungicides, and herbicides, as it is expected that most of the 'complete' dose-response curves will come from the first group of substances.

#### Data extraction

Mortality data at the single dose will be extracted. A tentative model for the data extraction is reported in the tables below.

**Table 10. Tentative data model for the data extraction (Study description)**

Parameter	Data type
Study ID	Key study identifier
Author	Free text
Source (DAR/dossier/Journal/Volume/Issue)	Free text
Guideline (if available)	Picklist <ul style="list-style-type: none"> <li>• OECD TG 213</li> <li>• OECD TG 214</li> <li>• OECD TG 246</li> <li>• OECD TG 247</li> <li>• OECD Guidance Document 239</li> <li>• None</li> <li>• Other (free text)</li> </ul>
Species	Picklist (to be implemented after preliminary analysis of availability)
Bee group	Picklist

	<ul style="list-style-type: none"> <li>• Honey bees</li> <li>• Bumble bees</li> <li>• Solitary bees</li> </ul>
Tested substance	Picklist (to be implemented after preliminary analysis of availability)
Tested substance group	Picklist <ul style="list-style-type: none"> <li>• Insecticide</li> <li>• Herbicide</li> <li>• Fungicide</li> </ul>
GLP	TRUE/FALSE
Endpoint time scale	Picklist <ul style="list-style-type: none"> <li>• Acute</li> <li>• Chronic</li> <li>• Other</li> </ul>
Test duration [days]	Float
Exposure type	Picklist <ul style="list-style-type: none"> <li>• Contact (direct application)</li> <li>• Contact with treated surface</li> <li>• Oral</li> <li>• Oversprayed</li> <li>• Other (free text)</li> </ul>

**Table 11. Tentative data model for the data extraction (Results)**

Dose	Integer (control marked as 0)
Dose Unit	Picklist <ul style="list-style-type: none"> <li>• µg/bee</li> <li>• µg/bee/day</li> <li>• µg/L</li> <li>• µg/kg</li> <li>• µg/cm<sup>2</sup></li> <li>• Other</li> </ul>
Tested bees at the dose	Integer
Affected bees at the dose	Integer
Measured Effect	Float [0-1]

### Data analysis and appraisal

Effects observed at each dose will be corrected for the effects observed in the control, so that for all studies mortality will be constrained between 0% and 100%. Study specific dose-response curves will be fitted, preferably using 2-parameters log-logistic models.

The monotonicity of the curve and the fitting will be assessed using appropriate statistics and via visual check of the corresponding plots.

The distribution of the slopes for the individual studies will be checked, and comparison with standard distributions (e.g. normal, log-normal, etc.) will be performed. If enough data will be available, comparisons between test durations, groups of bees, and groups of substances will be performed as well, to see whether appropriate extrapolation factors would need to be defined for each or whether some pooling is possible.

### 4.2.3. Suitability of the OECD 239 Guidance Document and the expected pesticide residue levels of the processed food of honey bees

#### 4.2.3.1. Background of the issue

In addition to pollen and nectar, certain honey bee life stages consume other food items, such as Worker Jelly or Royal Jelly which is fed to larvae and queens. After emerging from egg, honey bee larvae are provided with food produced by the worker bees (nurse). Larvae are fed with either Worker or Royal Jelly, depending on whether the larva is reared to develop in a worker bee or a queen. This jelly is a product of the hypopharyngeal and mandibular glands. The composition of Worker and Royal Jelly is similar in proteins, sugar and lipids within the first three days (Brouwers et al., 1987). Worker larvae are fed solely from pharyngeal gland secretions for the first three days, after which some honey/nectar and pollen are introduced into the diet (Shuel and Dixon, 1959). Queen larvae are fed only Royal Jelly throughout their whole development, and even after adult emergence, the food of the queen solely consists of Royal Jelly (Shuel and Dixon, 1959).

There is a discrepancy concerning the exposure in the test protocol for a honey bee chronic larval toxicity test according to OECD guidance document 239 on the one hand, and according to the EFSA, 2013 on the other hand. For the Tier 1 risk assessment presented in the EFSA, 2013, a toxicity endpoint for larvae derived from a chronic larval toxicity test with repeated exposure is required. An internationally agreed protocol for such a test was not yet available in 2013. Therefore, it was proposed in Appendix O of the EFSA, 2013 to use a protocol based on OECD test guideline 237 (larval toxicity test with single exposure, which was still in a draft version in 2013), and to dose larvae throughout their complete developmental period.

In 2016, an OECD guidance document was published for a honey bee larval toxicity test with repeated exposure (OECD guidance document 239), which has been used as the standard protocol for honey bee larval testing since. Within this test protocol, larvae are fed with uncontaminated food on Day 1 of the test and they do not get food on Day 2 (i.e. no exposure in the first two days of the development, in which the larvae are smallest and therefore potentially most sensitive). They receive food contaminated with the test chemical daily from Day 3 to Day 6. Consequently, exposure in a test according to this protocol (4 days) is shorter compared to what is recommended in the 2013 EFSA guidance document (exposure throughout the whole developmental period).

In OECD guidance document 239, it is not explained why the larvae are not exposed to the test substance in the first two days of the test. It should therefore be investigated whether the lack of exposure during the first two days is representative for what can be expected in the field. As larvae are fed only with jelly during the first days, this depends on the residues found in jelly.

Based on the above, the following question need to be answered:

#### **What is the magnitude of the residues in jelly compared to residues found in pollen and nectar?**

Answering the above questions would allow to determine to what extent there is a transfer of pesticide residues from nectar and pollen consumed by nurse bees to the jelly produced by these bees (thus the relative exposure level of the honey bee larvae in the first few days of their life, as well as the relative exposure level of the honey bee queen).

#### 4.2.3.2. Review process that was followed

A narrative search was performed in different databases (e.g. Google Scholar, ScienceDirect, SpringerLink) using search strings such as "residues in jelly", "pesticide residues in royal jelly" and "pesticide residues in worker jelly". In addition, publications referred to in any of the consulted articles, which contained potentially relevant information, were also assessed.

A number of publications were found that focused on potential contamination of royal jelly from the human health perspective (i.e. human consumption of jelly as a food supplement). In most royal jelly samples tested no residues above the LOQ (generally in the range of 0.25-20.0 µg/kg) were detected. Since these samples were from commercially available royal jelly, for which there is no information on the location of the hive from where they were collected, the usefulness of these data was considered as rather limited.

**Davis and Shuel (1988)** investigated the distribution of carbofuran and dimethoate in nurse bees, royal jelly and queen larvae. For this purpose, nurse bees that were fed with a glucose solution containing <sup>14</sup>C-labelled carbofuran or dimethoate at sub-lethal doses were dissected, and the radioactivity was measured in different organs and glands. In addition, sucrose syrup contaminated with <sup>14</sup>C-labelled carbofuran or dimethoate was provided as food to worker bees in small queenless units of about sixty bees. Twelve hours after the syrup was administered, two young larvae (1-2 days old) were grafted into beeswax queen cup and placed in the cages, so that the queenless units would have an opportunity to rear queens. After 72 hours, larvae and royal jelly was harvested from the queen cells and radioactivity was measured.

Within the nurse bees, very low levels of radioactivity were detected in the hypopharyngeal and mandibular glands. The majority of radioactivity was found in the digestive track (e.g. in the honey sac shortly after feeding, and later on also in the ventriculus and rectal sac). In larvae and larval food, <sup>14</sup>C-activity was detected for both substances tested, although at low levels. On a weight basis, concentrations of insecticide in the syrup provided to the nurse bees were  $5.45 \times 10^3$  (carbofuran) and  $3.33 \times 10^3$  (dimethoate) times higher than those in larval food. Based on these results, Davis and Shuel (1988) concluded that carbofuran and dimethoate are not secreted in significant quantities by the brood-food glands of nurse bees into larval food. The results of Davis and Shuel (1988) are in agreement with a similar experiment that was performed by Wittmann (1982 – as cited in Davis and Shuel, 1988, and Böhme et al., 2018a) with carbaryl and diflubenzuron.

**Böhme et al. (2018a)** performed an experiment in which the transfer from pesticide residues in the diet of worker bees to royal jelly fed to queen larvae was investigated. For that purpose, three queenless mini-colonies were set up, consisting of about 3500 bees. Two of the colonies were offered 70-g packages of a pollen-honey diet enriched with a mixture of pesticides that are commonly found in pollen pellets in intensively used agricultural areas in Southern Germany (high end field relevant concentrations). To avoid dilution of the pesticides by pollen collected in the field, a pollen grid was attached at the entrance. The third colony served as a control.

30 neonate larvae were grafted into plastic cell cups on a cell bar frame in each hive and after 2 days, the royal jelly were harvested and subjected to multi-pesticide residue analysis. This process was repeated three times in total.

Five pesticides with a comparatively higher limit of detection (boscalid, LOD = 2.67 µg/kg; dimethenamid-P, 1.33 µg/kg; methiocarb, LOD = 0.67 µg/kg; tebuconazole, LOD = 0.67 µg/kg; triadimenol, LOD = 1.33 µg/kg) were not detected in any of the samples from the exposed colonies. Due to matrix-related interferences, prothioconazole could not properly

evaluated. Thus, only seven of the 13 substances fed to the bees could be detected above their LOD. From all of the samples collected, pesticides could be detected above their limit of quantification (LOQ) in 17 cases. Concentrations of pesticides ranged from 0.42 and 2.16 µg/kg, with 76.5% of all pesticide concentration being below 1 µg/kg. Only in one sample could all seven pesticides be found. The insecticide thiacloprid was found in every sample, but only once in a concentration slightly above 2 µg/kg.

According to the authors, the percentage of pesticides potentially transferred from the offered food to royal jelly ranged between 0.001% (pyraclostrobin) and 0.016% (thiacloprid). These figures however could not be replicated by the working group. Based on the recalculation performed by the WG (that took into account only those pesticides for which residues above the LOQ were measured), these figures were in 0.07% to 0.45% (the concentration in the pollen-honey diet is a factor of 222 to 1404 higher compared to the concentration measured in royal jelly).

**DeGrandi-Hoffman et al. (2013)** and **Johnson and Percel (2013)** both performed similar experiments, which aimed at determining the effects of feeding on pesticide contaminated pollen on immature queen development, emergence and survival. In both studies, contaminated pollen (with single pesticide or with a combination of pesticides) were provided to small queenless colonies. Worker bees in the test colonies were confined, so that the diet and pesticide exposure of the bees during the experiment could be controlled. Young larvae were grafted in queen cells and were placed in the colonies to become new queens. In both studies, pesticide concentrations in the pollen fed to the test colonies and in the royal jelly harvested from queen cells were measured. DeGrandi-Hoffman et al. (2013) additionally measured concentrations in the worker bees.

In the study by DeGrandi-Hoffman et al. (2013), all three tested substances were detected in nurse bees, which was attributed to the presence of contaminated pollen in their gut. However, no residues were detected in royal jelly (noting that the LOQ of the analytical method was not reported in the paper). Despite the lack of residues in royal jelly, the emergence rate of queens was lower in the colonies fed with contaminated food compared to the control colonies. The authors attributed the lower queen emergence to a suppression of nurse bee vitality and/or immunity (which would result in a higher virus transfer to queen larvae).

In the study by Johnson and Percel (2013), different concentrations of the fungicides in pollen, which bracketed the fungicide concentrations observed in field-collected pollen, were tested. Residues in pollen were not measured directly after application, but after the pollen frames were kept in the hives for 4 days. The measured concentrations in pollen and royal jelly were only reported for the highest tested concentration of 400 mg/kg of a product which contained a nominal concentration of 101 mg/kg boscalid and 51 mg/kg pyraclostrobin. The measured concentrations in pollen after 4 days were  $47 \pm 5$  mg/kg boscalid and  $22 \pm 2$  mg/kg pyraclostrobin. Three of four royal jelly samples taken from this treatment contained detectable levels of pyraclostrobin (47-52 µg/kg), but boscalid was not detected in any royal jelly sample (again, the LOQ of the analytical method used was not reported). Based on these results, the concentration of residues in royal jelly were about a factor of at least 500 below the concentration in pollen. No effects on queen emergence and survival were observed in this study.

**Böhme et al. (2019)** performed an experiment in which the transfer from pesticide residues in the diet of worker bees to worker jelly fed to 3 to 6-day old worker larvae was

investigated. For that purpose, twenty mini-colonies, consisting of about 3500 bees and one mature queen were used. At the start of the experiment the sister queens were confined to an empty comb for 24h, to obtain larvae of defined age. From day three of the experiment, each colony was offered a 60-g package of a pollen-honey diet, which was replaced by a new package every second day. The amount of pollen-honey diet consumed was assessed daily. The pollen-honey diet of the non-control colonies was contaminated with a mixture of pesticides that are commonly found in pollen pellets in intensively used agricultural areas in Southern Germany. To avoid dilution of the pesticides by pollen collected from the field, a pollen grid was attached at the entrance.

The number of pollen grains and their botanical origin (palynology) in the worker jelly was determined. Further, the worker jelly samples were subjected to multi-pesticide residue analysis to determine the concentration of the different active substances.

In the worker jelly of three-day-old larvae, only 6 of the 13 substances offered were detected. In worker jelly of four-day-old larvae, already 10 substances were detectable, and in worker jelly of five- and six-day-old larvae twelve different pesticides were found (also the amount of harvestable jelly increased with the age of the larvae). The level of contamination also increased from 2.9-99.5 µg/kg in the worker jelly of the youngest larvae to 21.7-871.0 µg/kg in the oldest larvae. Concentrations of the active ingredients increased steadily with larval age.

The number of pollen grains also increased with increasing age for the larvae from 102.9 to 29533.7 pollen/cell accounting for 40.9 to 4653.5 pollen/mg worker jelly, respectively. Linear regression revealed a significant correlation between the increasing amount of pollen per mg worker jelly and the increasing pesticide concentrations.

The percentage of pesticides potentially transferred from the offered food to worker jelly ranged between 0.0009% and 0.0058%, 0.0001 and 0.0398%, 0.0037% and 0.0917%, and 0.0061 and 0.1478% for 3, 4, 5, and 6-day old larvae, respectively. The potential transfer rate of pesticides from the contaminated diet to royal jelly in the study by Böhme et al. (2018a) was comparable to the potential transfer rate to worker jelly of three- to four-day old larvae (based on the reported figures by the authors). For five- to six-day old larvae, the potential transfer rate to worker jelly was about 10 times higher. Taking into account the correlation between amount of pollen and pesticide concentrations, this was attributed to the pollen contained in the jelly. However, it should be noted that as for Böhme et al. (2018a), some values could not be replicated by the calculations of the WG (which adds some uncertainties to the outcome). Nevertheless,, the results of this study suggest that worker jelly is contaminated through residues in pollen incorporated into the jelly, rather than through transfer of residues through the glandular secretions.

#### Summary:

In the different studies where pesticide-contaminated pollen were provided to worker bees, only very low concentrations of these pesticides were found in the royal jelly produced by these worker bees. For example, for those substances which led to detectable residues in the study by Böhme et al. (2018a), the concentration in royal jelly was a factor of about 200 to 1400 below the concentration in the pollen-honey diet. Similarly, in the study by Johnson and Percel (2013), there was a difference of about a factor of 500 in the respective concentrations for pyraclostrobin. In the study by Davis and Shuel (1988), the concentration in the contaminated syrup provided to the nurse bees was also about three orders of magnitude higher compared to the residues in royal jelly.



When looking at residues in worker jelly of older larvae, Böhme et al. (2019) showed that the concentration of pesticide residues increased with larval age. Because of the correlation between the amount of pollen in the worker jelly and pesticide concentrations, it seems that worker jelly is contaminated through residues in pollen incorporated in the jelly, rather than through transfer of residues through the glandular secretions. With increasing larval age, sugars from nectar in the honey-sac are also increasingly added to the larval food (Brouwers et al., 1987). Therefore, contaminated nectar is likely also a source of increased pesticide concentrations in the worker jelly.

#### **4.2.3.3. Preliminary conclusions**

The data analyses as outlined above revealed that only a very small percentage of pesticide residues in pollen and nectar are transferred to the royal jelly (and worker jelly in the first days). Therefore, exposure of honey bee worker larvae during the first two days of their development is likely to be negligible. Consequently, the fact that larvae in a study according to OECD guidance document 239 are not exposed to contaminated food in the first two days of their development is not considered problematic. Therefore, an endpoint derived from such a study should be suitable for use in the risk assessment.

Also, exposure to queen larvae and emerged queens to pesticide residues through their food (royal jelly) is also considered to be very low to negligible. This does however not exclude any effects of pesticides on e.g. the emergence rate of queens, as shown by DeGrandi-Hoffman et al. (2013). It is however likely that other mechanisms than dietary exposure (e.g. suppression of nurse bee vitality and/or immunity) are responsible for these effects.

## PART 5

### 5. Planned methods for assessing high priority RA parameters or topics

#### 5.1. Methods to assess crop attractiveness

The revision of the list of attractive crops as defined in EFSA, 2013 is explicitly mentioned in the ToR 3 of the mandate. In view of this, of the sensitivity of the topic, and of the lack of consensus among the experts, the WG considered this task among the highest priorities for the revision.

ToR 3 is further divided for 3 sub-sections:

- Crop attractiveness for pollen and/or nectar
- Crop attractiveness for guttation fluids
- Consideration of harvesting time (before or after flowering)

Exposure via guttation water is addressed in section 4.1.6 and therefore is not further considered in the present section. This section on the other hand illustrates the methodology for addressing sub-points 1 and 3.

It is noted that crops can be attractive due to elements other than pollen, nectar, or guttation fluid, but these are not covered here.

##### 5.1.1. Definitions

Bees	The analysis is performed for the three bee groups, i.e. honey bees, bumble bees, and solitary bees
Nectar producing	A plant phenological stage of a crop is counted as "nectar producing", if the crop is flowering with nectar production, or the crop has extrafloral nectaries
Accessible nectar	Produced nectar is called "accessible", if at least one type of bees is able to collect the nectar
Pollen producing	A plant phenological stage of a crop is counted as "pollen producing", if the crop has pollen
Active collection of pollen	Produced pollen is called "active collected", if the pollen produced by a crop is collected by bees for food or to provision a colony or nest.

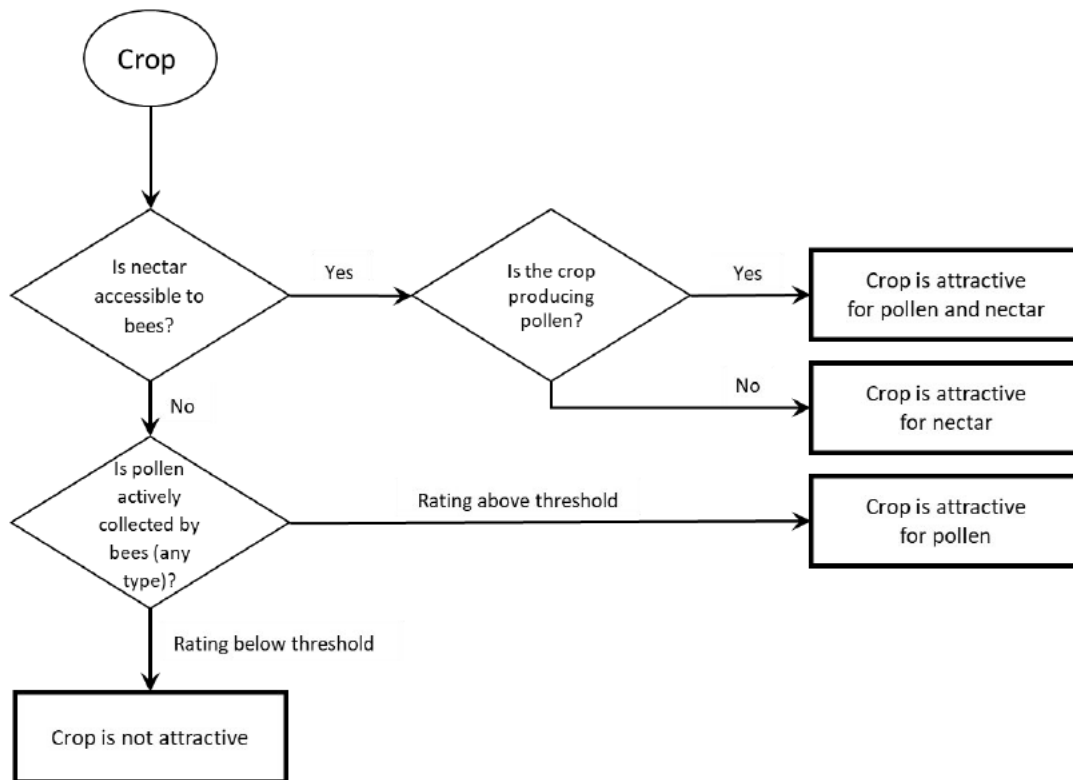
##### 5.1.2. Problem formulation and assessment model

To evaluate if a crop is attractive to bees, at least one of the following two criteria must be fulfilled:

1. The crop has at least one plant phenological life stage, where nectar is accessible to at least one type of bee
2. The crop has at least one plant phenological life stage, where pollen is actively collected by at least one type of bee

Please note that this definition does not quantify the attractivity of crops, esp. does not allow to compare crops on the level of their attractivity to bees (Crop A is more attractive than Crop B). Therefore, additional information on the quality of nectar and pollen is not needed.

The WG proposed a workflow to assess attractiveness based on the criteria above.



In order to go through this assessment scheme, the following information need to be considered:

Criterion	Description
Plant phenological life stage with nectar production (flowering, extrafloral nectaries)	e.g. BBCH codes
Does the agricultural practise allow a nectar producing stage?	yes/no, which practise (e.g. food, seed etc.), which stage (e.g. BBCH codes)
Is the nectar accessible to bee?	yes/no, eventually type of bees
Plant phenological life stage with pollen production	e.g. BBCH codes
Does the crop production allow a pollen producing stage?	yes/no, which practise (e.g. food, seed etc.), which stage (e.g. BBCH codes)
Is the produced pollen actively collected by any kind of bees?	<b>Expert rating</b> , see operationalisation of the EKE question
Attractiveness	yes/no

As reported in the table above, after evaluating the retrieved evidence on “active collection of pollen by bees”, the WG considered that the information available in the literature would not be sufficient to address the issue. Hence, the WG agreed that a formalised Expert Knowledge Elicitation (hereafter EKE) would be the best way forward. The draft methodology for this EKE follows the principles of the EFSA Guidance on Expert Knowledge in Food and Feed Safety Assessment (EFSA, 2014).

Apart from the identified need of an Expert Knowledge elicitation, reported above, the draft methodology – included in Appendix B – reports all other relevant settings of an EKE:

- the elicitation question
- the expert profiles
- the elicitation methodology to be applied

Changes of the methodology proposed in this protocol may occur as result of the review process, during the final selection of the group of experts, and during the elicitation itself. All changes will be documented together with the results of the elicitation.

### 5.1.3. Crop list

As preparation of the Expert Knowledge Elicitation the WG on Bee Guidance Revision will prepare a complete list of possible attractive crops (genus, species, cultivars).

Crops are in the first step evaluated on genus level. Some exceptions are given:

- In case not all species and/or cultivars of the genus will get the same evaluation result appropriate groups of species are defined case by case
- In case different agricultural practises will lead to different evaluation results the crop is specified also by the agricultural practise, e.g. crop for food, crop for seed production. Especially, if the for food production the crop is harvested before flowering, it will be evaluated, if the crop is used for seed production.

**Table 12.** The evaluation will include the following aspects of the assessment. The evaluation follows the scheme above and ends with the statement on attractiveness for nectar and/or pollen. List of crops with criteria

Genus	Species / cultivars, if needed	Agricultural practise, if needed	Plant phenological life stage with nectar production	Does the agricultural practise allow a nectar producing stage	Is the produced nectar accessible to bees	Plant phenological life stage with pollen production	Does the crop production allow a pollen producing stage	Is the produced pollen actively collected by any type of bees
								To be filled after the EKE

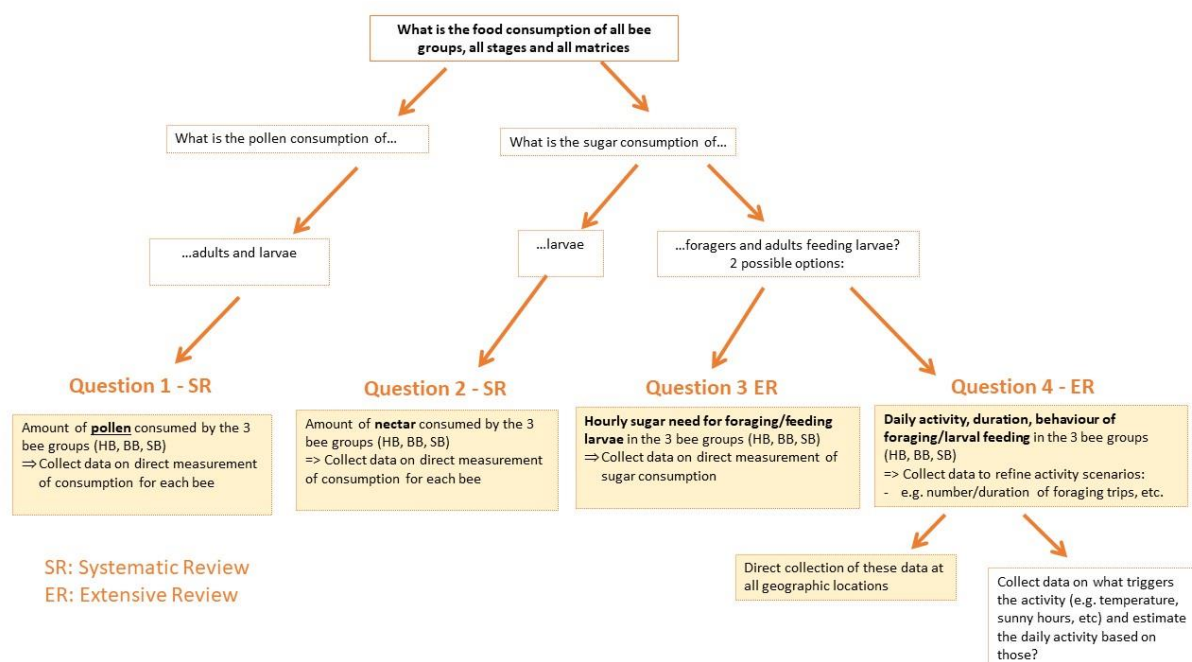
## 5.2. Methods to estimate food consumption: systematic review

Considering the outcome of the sensitivity analysis (Appendix A) and that for the EFSA, 2013 no systematic data collection was applied, the food consumption was considered by the guidance revision WG as a high-priority parameter. Therefore, a systematic review is planned.

### 5.2.1. Question formulation

The question 'food consumption' was further elaborated and divided into four main questions (Qs) based on the rationale depicted in Figure 4.

- Q1: What is the **amount of pollen** consumed by the different bee species (adults and larvae) native to Europe? This question will be answered via SR.
- Q2: What is the **amount of sugar** consumed by the larvae of different bee species native to Europe? This question will be answered via SR.
- Q3: What is the **hourly sugar need** due to the different **activities** (e.g. foraging and nursing activities) of the different adult forms of the different bee species native to Europe? This question will be answered by identifying relevant studies through an extensive literature search followed by evidence screening based on pre-defined eligibility criteria; then, studies will be appraised and synthesised in a narrative manner. This intermediate methodological choice is because it is not expected that the new data collection will result in considerable different outcome compared to the existing knowledge as in the current guidance.
- Q4: What is the **daily duration of the foraging and larval feeding behaviours** of the different adult forms of the different bee species native to Europe? The approach and rationale for this choice is the same as for the previous question.



**Figure 4. Rationale for formulating the review questions on food consumption**

### 5.2.2. Eligibility criteria for study selection

The criteria for selecting studies for inclusion in these reviews are reported in the tables below. They will be pilot tested on a subset of records and refined if prone to misinterpretation by the reviewers.

**Table 13. Criteria for selecting studies based on study characteristics, for question 1**

<b>Study design</b>	<b>In</b>	<ul style="list-style-type: none"> <li>- Studies which measure the <b>amount of pollen</b> consumed by individual bees (for honey bees: nurses; for other bee species: worker, queen and drone larvae).</li> <li>- Dissection studies to assess pollen gut content of larvae.</li> <li>- Lab. studies to assess pollen consumption by nurses with the development of hypopharyngeal glands (HPGs) that mimic the normal development of the HPGs.</li> <li>- Lab. studies to measure the pollen consumption with full control on the consumed food (e.g. no access to alternative food sources).</li> <li>- Greenhouse/semi-field studies measuring larval consumption (pollen) of solitary bees.</li> </ul> <p>[Semi-field studies = studies with a limited foraging area with attractive crops to bees delineated by a net/shelter that will not be influenced by external pedoclimatic conditions]</p>
	<b>Out</b>	<ul style="list-style-type: none"> <li>- Standard toxicity test studies (e.g. OECD),</li> <li>- Studies with access to alternative food sources (with no measurement of this food).</li> <li>- Greenhouse and field studies assessing pollen consumption at colony level – for honey bees and bumble bees (which do not relate to individual adult consumption <i>vs.</i> individual larval consumption).</li> </ul> <p>[All of these study typologies are not considered to sufficiently mimic realistic pollen consumption by bees under their normal daily activities or these studies are not considered specific enough to determine individual consumption]</p>
<b>Population</b>	<b>In</b>	Honey bees ( <i>Apis mellifera</i> ) Bumble bees ( <i>Bombus</i> spp.) Solitary bees All bee types living in the EU
	<b>Out</b>	Bee species that are not native in Europe

<b>Outcome</b>	<b>In</b>	Amount of pollen consumed by: <ul style="list-style-type: none"> <li>- honey bees: nurses and larvae (worker, queen, drone)</li> <li>- bumble bees: adults (workers/founding queen) and larvae (worker, queen, drone)</li> <li>- solitary bees: adults and larvae (female and male)</li> </ul>
	<b>Out</b>	Amount of pollen (in terms of types/species diversity) consumed by bees. Amount of pollen collected and/or consumed at colony level.
<b>Method</b>	<b>In</b>	All possible methods
	<b>Out</b>	Studies dealing with pollen traps and palynological analysis (to assess food consumption in terms of pollen diversity or to assess foraging preferences)
<b>Location</b>	<b>In</b>	All possible locations
	<b>Out</b>	/

**Table 14. Criteria for selecting studies based on study characteristics, for question 2**

<b>Study design</b>	<b>In</b>	<ul style="list-style-type: none"> <li>- Studies which measure the <b>amount of nectar</b>, and the sugar content in nectar, consumed by individual larvae (for honey bees: workers, queen and drones; for bumble bees: workers and founding queen; for solitary bees : females and males).</li> <li>- Studies which determine the amount of <b>sugar/carbohydrates</b> required by larvae (for honey bees: workers, queen and drones; for bumble bees: workers and founding queen; for solitary bees : females and males).</li> <li>- Greenhouse/semi-field studies measuring larval consumption (nectar) of solitary bees.</li> </ul> <p>[Semi-field studies = studies with a limited foraging area with attractive crops to bees delineated by a net/shelter that will not be influenced by external pedoclimatic conditions]</p>
	<b>Out</b>	<ul style="list-style-type: none"> <li>- Standard toxicity test studies (e.g. OECD).</li> <li>- Field studies assessing nectar consumption at colony level – for honey bees and bumble bees (which do not relate to individual adult consumption vs. individual larval consumption).</li> </ul> <p>[All of these study typologies are not considered to sufficiently mimic realistic sugar consumption by larvae under their normal development or these studies are not considered specific enough to determine individual consumption]</p>

<b>Population</b>	<b>In</b>	Honey bees ( <i>Apis mellifera</i> ) Bumble bees ( <i>Bombus</i> spp.) Solitary bees all bee types living in the EU
	<b>Out</b>	Bee species that are not native in Europe
<b>Outcome</b>	<b>In</b>	Amount of nectar/sugar/carbohydrates consumed by larvae: <ul style="list-style-type: none"> <li>- honey bees: queen, worker and drone</li> <li>- bumble bees: founding queen, worker and drone</li> <li>- solitary bees: male and female</li> </ul>
	<b>Out</b>	Amount of honey collected and/or consumed by colonies (in honey bees/bumble bees)
<b>Location</b>	<b>In</b>	All possible locations
	<b>Out</b>	/

**Table 15. Criteria for selecting studies based on study characteristics, for question 3**

<b>Study design</b>	<b>In</b>	Lab. studies which are based on measurement of energetic needs (e.g. kJ/h) for individual foraging bees (e.g. with flying arena) and nurses.
	<b>Out</b>	Any lab studies which do not provide information on energetic needs (e.g. kJ/h) for individual foraging bees (e.g. with flying arena) and nurses.
<b>Population</b>	<b>In</b>	Honey bees ( <i>Apis mellifera</i> ) Bumble bees ( <i>Bombus</i> spp.) Solitary bees all bee types living in the EU
	<b>Out</b>	Bee species that are not native in Europe
<b>Outcome</b>	<b>In</b>	Amount of energy required by bees to forage in the landscape (honey bees, bumble bees and solitary bees)  Amount of energy required by nurses to perform their daily activity (honey bees, bumble bees and solitary bees)
	<b>Out</b>	/
<b>Location</b>	<b>In</b>	All possible locations
	<b>Out</b>	/



**Table 16. Criteria for selecting studies based on study characteristics, for question 4**

<b>Study design</b>	<b>In</b>	<p>- Field studies on the <b>daily activity of foragers</b> (i.e. activity <i>versus</i> resting in a day) for all bee species (honey bees, bumble bees and solitary bees) under different environmental conditions (season, climate/weather, landscape) from EU.</p> <p>- Field studies on the <b>foraging activity</b> (i.e. flying <i>versus</i> searching food <i>versus</i> collecting food within one foraging return trip) for all bee species (honey bees, bumble bees and solitary bees) under different environmental conditions (season, climate/weather, landscape) from EU</p> <p>- Field studies on <b>foraging behaviour</b> in bees (e.g. number and duration of foraging trips) under different environmental conditions from EU (season, climate/weather, landscape)</p> <p>- Field and semi-field studies with hives/colonies on the <b>daily activity of the nurses</b> (honey bees and bumble bees) under different environmental conditions (season, climate/weather, landscape) from EU</p> <p>- Greenhouse/semi-field/field studies on the <b>daily activity of the solitary bees</b> feeding their brood</p> <p>[Semi-field studies = studies with a limited foraging area with attractive crops to bees delineated by a net/shelter that will not be influenced by external pedoclimatic conditions]</p>
	<b>Out</b>	<ul style="list-style-type: none"> <li>- Lab studies cannot mimic the normal activity of the foraging bees in the natural conditions (reflecting the inherent variability of the environment)</li> <li>- Standard toxicity test studies (e.g. OECD)</li> <li>- Cognitive tests (e.g. PER, labyrinth)</li> <li>- Semi-field studies where colony is artificially altered (e.g. micro-colonies where number of adults or brood frames removed/reduced)</li> </ul>
<b>Population</b>	<b>In</b>	<p>Honey bees (<i>Apis mellifera</i>)</p> <p>Bumble bees (<i>Bombus</i> spp.)</p> <p>Solitary bees</p> <p>all bee types living in the EU</p>
	<b>Out</b>	Bee species that are not native in Europe
<b>Outcome</b>	<b>In</b>	<ul style="list-style-type: none"> <li>- Daily activity of foragers (foraging/resting)</li> <li>- Foraging activity per trip (flying/searching/collecting food)</li> <li>- Foraging behaviour (number and duration of flying trips per day)</li> <li>- Daily activity of bees feeding larvae</li> </ul> <p>All the above is recorded under specific environmental conditions (seasons, climates, landscapes/resources and</p>

		weather) in EU
	<b>Out</b>	/
<b>Location</b>	<b>In</b>	All possible locations
	<b>Out</b>	/

**Table 17. Criteria for selecting studies related to report characteristics**

<b>Time</b>	<b>In</b>	No time limits
	<b>Out</b>	/
<b>Language</b>	<b>In</b>	European languages (with abstract in English)
	<b>Out</b>	Rest of languages
<b>Publication type</b>	<b>In</b>	<ul style="list-style-type: none"> <li>• Primary research studies (i.e. studies generating new data)</li> <li>• Conference abstracts or posters if they contain primary data.</li> <li>• PhD theses and dissertations</li> </ul> <p>Reviews will be used as sources of further references and to assess the appropriateness of the search strategy applied</p>
	<b>Out</b>	<ul style="list-style-type: none"> <li>• Letters to editor</li> <li>• Expert opinions</li> <li>• Editorials</li> </ul>

### 5.2.3. Search strategies

The bibliographic databases listed in Table 18 will be searched to identify relevant studies. The databases selected have been identified in line with the defined scope of the review.

**Table 18. Bibliographic databases searched for relevant studies**

<b>Source of information</b>	<b>Platform</b>
BIOSIS Citation Index (1926-present)	Web of Science
CAB Abstracts (1920-present)	Web of Science
Scopus (inception-present)	Scopus
Zoological Record (1864-present)	Web of Science
Web of Science Core Collection <ul style="list-style-type: none"> <li>• Science Citation Index Expanded (1975-present)</li> <li>• Conference Proceedings Citation Index- Science (1990-present)</li> <li>• Emerging Sources Citation Index (2005-present)</li> </ul>	Web of Science
DART-Europe E-theses Portal	DART Europe
EBSCO Open Dissertations	EBSCOhost

The search strings that will be used to capture relevant studies to the review question are presented in Appendix C.

Three different search strategies have been designed to retrieve the evidence to answer the four questions. All of them contain terms to identify the population bee species (population) and a) amount of pollen, nectar, sugar or carbohydrates for Q1 and Q2, b) energy consumption for Q3 or c) flying activity, foraging behaviour for Q4.

A wide range of search terms are used to cover language variations (synonyms, related terms, lay and scientific terminology, etc.). Several sources have been used to select the search terms: studies included in (EFSA, 2013); thesaurus such as, Biosis Citation Index, CAB Thesaurus and Zoological Record Thesaurus; the European Red List of Bees (Nieto et al., 2014) has been consulted in order to gather all the bee species of interest. The terms have been combined using the appropriate Boolean and proximity operators.

The search is not limited by date. The language of the original studies will be limited to European languages for bibliographic databases, whereas the retrieval of Ph.D. theses will be limited to English.

The output of the searches will be loaded into Endnote bibliographic management software (Clarivate Analytics). Duplicate references will be removed by a combination of automatic and manual detection of duplicates.

The final search processes and strategies will be documented and reported in the technical report, i.e. the date of the search, sources of information, search string for each bibliographic database and additional sources, and the number of records before and after de-duplication. Should modifications in the search strings be considered after the publication of the protocol, we will report them.

#### 5.2.4. Study selection process

The records retrieved via the literature searches will be screened against the eligibility criteria defined above.

The study selection process will be carried out in two steps:

1. Step 1: titles and abstracts screening, to exclude obviously irrelevant records. All other apparently relevant or of unclear relevance records will be moved to the following step;
2. Step 2: full-text screening, to select records for inclusion/exclusion.

Each record will be screened by two independent reviewers to minimise the risk of error using DistillerSR® (Evidence Partners, Ottawa, Canada). Between-reviewer conflicts not solvable via discussion will be discussed at among all reviewers. If needed, the DistillerSR Artificial Intelligence (AI) function will be used as second reviewer at title and abstract screening, to speed up the selection process.

During the study selection process, studies published in multiple publications will be identified and duplicates removed. If needed, papers with abstract in English and full-text in another European language that seem relevant based on the abstract will be translated.

The results of the different phases of the record selection process will be reported in a flowchart as recommended in the PRISMA statement on preferred reporting items for systematic reviews and meta-analyses (Moher et al., 2009).

### 5.2.5. Evidence appraisal

For questions 1 and 2 (Figure 4), in this step of the process the Risk of internal and external Bias (RoB) of each included study will be assessed, separately.

In a descriptive study, a biased study is one that does not give a true representation of the situation to be described.

Internal bias (or limitation in internal validity) refers to any error in the conduct of the study that results in a conclusion which is different from the truth we are interested in. The method for measuring sugar content not being reliable/accurate is an example of source of internal bias in the studies relevant to this assessment.

External bias (or limitation in external validity) affects the extent to which the study results are generalisable to the assessment question, e.g. when the study settings are not being representative of the reference population/conditions/landscape settings.

The possible imprecision of the studies included in the assessment, which is related to random error and indicates the ability of a study to provide similar results when repeated under the same conditions, will be considered when synthesising the results (next section)

For each study addressing questions 1 and 2 (Figure 4), the appraisal will be done at the **outcome (gut content, HPG size, etc.) level**, because for the same study, the design and conduct may affect the RoB differently depending on the endpoints measured. The method for measuring sugar content in a study is an example of a potential source of bias that requires specific considerations depending on the endpoint under consideration. Internal and external validity (or risk of internal and external bias) will be appraised for each individual study using the critical appraisal tools (CAT) illustrated in the following tables. Each appraisal question is accompanied by a rationale for answering it, to make the appraisal as structured and transparent as possible and reduce the risk of subjectivity. The tool will be translated in the review management software DistillerSR® to allow web-based appraisal of the studies and facilitate the reporting of the results. Each study will be appraised by two independent reviewers. Possible discrepancies not solvable via discussion between the two reviewers will be discussed by the whole WG. If upon further discussion the WG cannot reach an agreement on a rating, the more conservative judgment (the highest RoB) will be selected. The CATs will be pilot tested by two reviewers. Feedbacks from this testing phase will be used for further refining this process. Hence, the CAT finally used for the assessment may present some modifications compared to the one reported here.

The studies addressing questions 3 and 4 in Figure 4 will be assessed in more narrative, traditional manner, that is, without pre-defining and applying any critical appraisal tools. However, some elements that will be considered for evidence appraisal can be anticipated. For instance, the temporal dimension of the seasonality of the bees (winter *versus* spring-summer) will be used for assessing external validity. For instance, if a study is carried out outside the EU, but in a geographical area with bio-climatic conditions comparable to at least one of the EU biogeographic regions, then the risk of the study results not being generalisable will be deemed probably low.

**Table 19. Critical Appraisal Tool for assessing the risk of internal bias (internal validity) of the studies included in the assessment**

Appraisal question (internal bias)	Rating	Rationale for answering the question	Relevance by assessment question and related outcomes	
			Q1	Q2
1. Are the test organisms free from unrealistic/unusual stressors affecting their feeding behaviour and energetic demands (e.g. not exposed to high levels of infectious agents and/or pesticide residues in food which are likely to alter their behaviour)?	Definitely low RoB	The tested organisms are known to be healthy (or with realistic disease prevalence) and immune from previous stress and not exposed to any chemicals which are likely to alter their behaviour.		
	Probably low RoB	No specific information is reported but the methodology raises no particular concern for disease/chemical exposure or unrealistic stress.		
	Probably high RoB/NR	Some indications are available suggesting that the tested bees might have been stressed before or during the experiment or whether they might be affected by diseases with an unrealistic prevalence. OR Some indications are available suggesting that the tested bees might have been stressed before or during the experiment or whether they might be affected by chemical exposure, OR No suitable information for assessing the health or the stress status of the bees is available.	X	X
	Definitively high RoB	It is known that the tested bees were unrealistically stressed before or during the experiment AND/OR Bees were known to be affected by some disease with an unrealistic prevalence. AND/OR		

Appraisal question (internal bias)	Rating	Rationale for answering the question	Relevance by assessment question and related outcomes	
			Q1	Q2
		Bees were known to have their behaviour altered by chemical exposure.		
<p>2. Is the methodology for measuring the relevant outcome (endpoint) accurate (there are different methodologies to assess food consumption, gut content, HPG, etc)?</p> <p>Note: this is a key appraisal question, i.e. a question that has a higher impact on the overall internal validity of a study for a specific outcome/endpoint</p>	Definitely low RoB	The method for measuring the outcome is well detailed and it is considered accurate to get a reliable estimation of the outcome in the conditions of the study. Ideally, an estimation of the accuracy of the method is given and it is satisfactory. The measuring methodology is unlikely to introduce behavioural changes in bees that may bias the outcome.		
	Probably low RoB	<p>The method for measuring the outcome is described in general terms but raise no concern about the reliability of the measurement.</p> <p><b>OR</b></p> <p>The method for measuring the outcome presents only minor issues in terms of accuracy.</p> <p><b>OR</b></p> <p>The method for measuring the outcome has potential for altering the bee behaviour, but the impact on the final outcome is considered low.</p>	X	X
	Probably high RoB/NR	<p>The method for measuring the outcome is not sufficiently described.</p> <p><b>OR</b></p> <p>No estimation about the accuracy of the method is given, and there are valid concerns that the method may not</p>		

Appraisal question (internal bias)	Rating	Rationale for answering the question	Relevance by assessment question and related outcomes	
			Q1	Q2
		provide an accurate estimation of the outcome. <b>OR</b> The method has the potential for inducing some behavioural changes with potential repercussion on the final outcome measurement.		
	Definitively high RoB	The methodology for measuring the outcome is known to be poorly accurate <b>OR</b> The method is likely to induce serious behavioural changes with repercussion on the final outcome measurement.		
3. Are food consumption measured at the specific level (i.e. per adult bee/day)?	Definitely low RoB	Measurements are conducted on individual bees per day	X for nurses (HB, BB)	NR larvae are assessed at the individual level and over one or several days of development
	Probably low RoB	Measurements are conducted on individual bees during a given number of days		
	Probably high RoB/NR	Measurements are conducted on a given number of bees (> 1 individual) over 1 day (i.e. assessments are for « n » bees per day)		
	Definitively high RoB	Measurements are conducted on a given number of bees (> 1 individual) and during a given number of days (> 1 day) (i.e. assessments are for « n » bee over « n » days)		
4. Is statistical analysis appropriate and consistent with the initial proposal of the study?	Definitely low RoB	There is clear evidence that the statistical analysis was appropriate <b>OR</b> The elaboration was wrong/inappropriate, but data are	X	X

Appraisal question (internal bias)	Rating	Rationale for answering the question	Relevance by assessment question and related outcomes	
			Q1	Q2
<p>(applicable only when the statistical results of the study are used. If raw data are provided, the statistical analysis will not be assessed as the data will be analysed separately)</p> <p>Note: It is expected that in very rare cases, some statistical analysis other than basic statistics (e.g. mean and SD) will be available (without the availability of the raw data) and will have to be appraised</p>		available with sufficient level of disaggregation (e.g. raw data, detailed table or plots) that allows performing a more appropriate analysis*		
	Probably low RoB	Data elaboration is available, although not all passages are clear from the reporting of the methodology. However, the degree of uncertainty is not likely to impair the reliability of the outcome analysis.		
	Probably high RoB/NR	Data elaboration is obscure and there are severe doubts that the presented outcome was estimated correctly. Underlying data not available.		
	Definitively high RoB	From the reporting in the methodology section it is clear that major errors were performed and/or inappropriate elaboration methods have been used for estimating the outcome. Underlying data not available.		



**Table 20. Critical Appraisal Tool for assessing the risk of external bias (external validity) of the studies included in the assessment. The tool covers one appraisal question only**

Appraisal question (external bias)	Rating	Rationale for answering the question	Relevance by assessment question and related outcomes	
			Q1	Q2
What is the confidence in the measured outcome for reliably estimating food consumption (e.g. proxies of food consumption such as gut content, HPG, etc.)?	Definitely low RoB	The measured outcome can be directly related to food consumption: a reliable estimation of the outcome gives immediately a reliable estimation of the food consumption.	X	X
	Probably low RoB	The measured outcome is logically related with the food consumption, but there are some uncertainties in extrapolating from one to the other. [For example, gut content is related to food intake, but the food may not be entirely digested/absorbed by the bee]		
	Probably high RoB/NR	The measured outcome is logically related with the food consumption, but there are major uncertainties in extrapolating from one to the other. [For example, the increase size of the HPG is related to food consumption below the average duration of the development of the HPG, i.e. < 10 days]		
	Definitively high RoB	The measured outcome is unlikely to offer a reliable picture of the food consumption. [For example, the increase size of the HPG is related to food consumption just over a few days which is far below the average duration of the development of the HPG]		

### 5.2.5.1. Summary of the internal validity and external validity of each individual study, by endpoint

For internal validity only one key appraisal question is identified (i.e. the question on the accuracy of the method for assessing the endpoint) (Table 21). The answers to the key and non-key appraisal questions will be combined into a single scoring method, classifying each endpoint from each study into a different tier reflecting the Risk of internal Bias, as explained in Table 22.

**Table 21. Summary of appraisal questions on internal validity.**

Appraisal questions assessing internal validity	Key (Y/N)
1. Are the test organisms free from unrealistic/unusual stressors affecting their feeding behaviour and energetic demands	N
2. Is the methodology for measuring the relevant outcome (endpoint) accurate?	Y
3. Are dietary exposures measured at the specific level (i.e. per adult bee and per day)? (relevant in some situations only)	N
4. Is statistical analysis appropriate and consistent with the initial proposal of the study? (relevant in some situations only)	N

**Table 22. Algorithm to combine the answers to the appraisal questions and allocate studies to tiers of Risk of internal Bias**

	Tier
<b>TIER 1: All appraisal questions (the key question and the non-key questions*) are scored ++/+ (definitively or probably low RoB)</b>	1 (low RoB)
*non-key appraisal questions are not relevant for some studies	
<b>TIER 2: study does not meet criteria for TIER 1 or TIER 3</b>	2
<b>TIER 3: the key appraisal question is scored – or --, regardless of the answer to the other questions</b>	3 (high RoB)

For the quantification of external validity, as only one appraisal question was identified for each aspect, there will not be the need to summarise. The results of critical appraisal will be reported for each individual study/endpoint along with the rationale for the rating and across the body of evidence using heatmap tables as for example the one reported in Table 23.

**Table 23. Example of heatmap for presenting the results of evidence appraisal**

Reference metadata	Endpoint(s)	Results	Internal validity (summarised across appraisal questions)	External Validity (based on one appraisal question only)
				Q2. what is the

			Tier 1	PHRoB
			Tier 3	DHRoB
			Tier 2	PHRoB

PHRoB: probably high risk of bias; DH risk of bias

### 5.2.6. Data extraction from the included studies and evidence synthesis

Studies may be retained for or discarded from the synthesis depending on the results of critical appraisal. If retained for the synthesis, studies will undergo a structured data extraction based on pre-defined data models and related instructions in DistillerSR®. Before implementation, the data extraction model will be pilot tested on a subset of studies and refined accordingly.

The present analysis attempts to provide answers to four different assessment questions, some of which present different internal strata, e.g. answers to Q1 will need to be specific for at least each of the three bee groups and different for adult and larvae. These different strata, together with further elements (e.g. characterisation of bee ages, spatial-temporal characterisation, where applicable) will have to be considered in the data extraction phase. In addition, it is expected that the answer to some assessment questions can be informed by heterogeneous outcomes (i.e. different outcome variables). The strategy for integrating heterogeneous outcomes will depend on the relative diversity, abundance, and validity of each contributing outcome. Hence, it is likely that preliminary analyses comparing results obtained from the different outcome variables will be performed before any integration.

It is further envisaged that, if enough valid information will be retrieved, an integration of the answers to Q3 and Q4 will be performed, in order to provide reliable estimates of the sugar intake for adult bees.

Considering the nature of the outcome variable of interest, a generalized linear mixed model could be used to estimate sugar content by crop accounting for other factors, such as landscape, environmental conditions, etc. The final choice of the method for synthesising evidence will depend on the granularity and heterogeneity of the studies retrieved.

If all studies are retained for the synthesis and they present different degrees of (internal and external) validity, subgroup analyses may be carried out to explore the impact on the final estimation of the parameter of interest. If subgroup analyses lead to different conclusions, an expert knowledge elicitation might be considered to reach an overall conclusion and characterise the related uncertainties. It is anticipated that depending on the amount, validity and heterogeneity of the available evidence, it may not be possible to conduct quantitative syntheses.

### 5.3. Methods to estimate crop-specific sugar content in nectar: systematic review

Sugar content in nectar is crop-dependent (genetics) and varies a lot due to abiotic factors like e.g. air humidity. For honey bees for instance, the sugar content in the nectar collected can range from 15 to 65%, while solitary bees can collect nectar with 10% of sugar (section 3.1 of Appendix J of EFSA, 2013). EFSA, 2013 guidance considered the worst case of 15% for honey bees. However, the WG agreed that nectar collection with these default sugar contents (15% or 10%) is likely to represent a rather rare situation. Therefore, for the revised guidance it was decided to further investigate this high-priority aspect by conducting a systematic review of the available evidence.

#### 5.3.1. Question formulation

The question that will be answered through the systematic review can be summarised as follows: 'What is the sugar content of the nectar of the different crops grown in the EU?'

The WG have considered to address the same question for the most abundant weed species occurring in agricultural areas in EU. However, finally the WG considered that the default value considered by the current guidance document (30% sugar content) is a realistic worst-case estimation for the weed scenario and field margin scenario that represents habitats that include simultaneously a number of taxa. Therefore, the question only for the crop species will be addressed.

#### 5.3.2. Eligibility criteria for study selection

The criteria for selecting studies for inclusion in the review on sugar content for crops are reported in Tables 24 and 25. They will be pilot tested on a subset of records and refined if prone to misinterpretation by the reviewers.

**Table 24. Criteria for selecting studies based on study characteristics**

<b>Study design</b>	<b>In</b>	Field studies, (i.e. studies performed in open settings where the variables naturally occur with minimal direct influence of human)  Semi-field studies (i.e. studies where the cultivated plant is protected by a net or any shelter that will not influence the pedoclimatic conditions)  Greenhouse studies (i.e. studies where the cultivated plant is protected by a shelter (usually plastic or glass))
	<b>Out</b>	Lab studies
<b>Population</b>	<b>In</b>	Crops grown in the EU and producing nectar (a list of crops is included in Appendix D of EFSA, 2013)
	<b>Out</b>	Crops not grown in the EU or that do not produce nectar
<b>Outcome</b>	<b>In</b>	Quantification of the sugar content of the nectar of crop species
	<b>Out</b>	Information which is not quantity/concentration of sugar in/from nectar and sugar content cannot be inferred even

		indirectly (e.g. by calculations/estimations)
<b>Sampling method</b>	<b>In</b>	<ul style="list-style-type: none"> <li>• Sampling directly the flowers: e.g. micro-capillary (microcaps, pipettes, syringes), centrifugation, discharged from the flower, using filter papers, using paper wicks, rinsed out, washed out</li> <li>• Derivation not directly from the flower: honey sack from bees caught during foraging in confined study set (semi field, greenhouse) or in the middle of the cropped field</li> </ul>
	<b>Out</b>	Honey sack from bees caught outside of the field (e.g. in front of the hive, edge of the field), from comb
<b>Method for determination of the sugar content</b>	<b>In</b>	All methods for sugar determination from polar matrix (e.g. refractometry, HPLC, spectrometry)
	<b>Out</b>	/
<b>Location</b>	<b>In</b>	All possible locations
	<b>Out</b>	/

**Table 25. Criteria for selecting studies related to report characteristics**

<b>Time</b>	<b>In</b>	No time limits
	<b>Out</b>	/
<b>Language</b>	<b>In</b>	European languages (with abstract in English)
	<b>Out</b>	Rest of languages
<b>Publication type</b>	<b>In</b>	<ul style="list-style-type: none"> <li>• Primary research studies (i.e. studies generating new data)</li> <li>• Conference abstracts or posters if they contain primary data.</li> <li>• PhD theses and dissertations</li> </ul> <p>Reviews will be used as sources of further references and to assess the appropriateness of the search strategy applied</p>
	<b>Out</b>	<ul style="list-style-type: none"> <li>• Letters to editor</li> <li>• Expert opinions</li> <li>• Editorials</li> </ul>

### 5.3.3. Search strategy

The bibliographic databases listed in Table 26 will be searched to identify relevant studies. The databases selected have been identified in line with the defined scope of the review.

**Table 26. Bibliographic databases searched for relevant studies**

<b>Source of information</b>	<b>Platform</b>
BIOSIS Citation Index (1926-present)	Web of Science
CAB Abstracts (1920-present)	Web of Science
Scopus (inception-present)	Scopus
Zoological Record (1864-present)	Web of Science
Web of Science Core Collection <ul style="list-style-type: none"> <li>• Science Citation Index Expanded (1975-present)</li> </ul>	Web of Science

- Conference Proceedings Citation Index- Science (1990-present)
- Emerging Sources Citation Index (2005-present)D

DART-Europe E-theses Portal	DART Europe
EBSCO Open Dissertations	EBSCOhost

The search strings that will be used to capture studies relevant to the review question are presented in Appendix D.

The search strategy comprises two concepts: a) sugar content in nectar (outcome) and b) crops grown in the EU producing nectar or bees (population). Terms for bees have been added to the population concept to identify studies sampling bees to assess the nectar content of a crop.

A wide range of search terms are used to cover language variations (synonyms, related terms, lay and scientific terminology, etc.). The previous EFSA guidance document on the risk assessment of plant protection products on bees (EFSA, 2013) has been used to identify crops producing nectar growing in the EU, this concept has been complemented with generic terms for crop, flowers, and fruits to be as extensive as possible.

The search is not limited by date. The language of the original studies will be limited to European languages for bibliographic databases, whereas the retrieval of Ph.D. theses will be limited to English.

The output of the searches will be loaded into Endnote bibliographic management software (Clarivate Analytics). Duplicate references will be removed by a combination of automatic and manual detection of duplicates.

The final search processes and strategies will be documented and reported in the technical report, i.e. the date of the search, sources of information, search string for each bibliographic database and additional sources, and the number of records before and after de-duplication. Should modifications in the search strings be considered after the publication of the protocol, we will report them.

#### 5.3.4. Study selection process

Same process as for this step in the systematic review on food consumption (see relevant section above).

#### 5.3.5. Evidence appraisal

Same introduction to the process for appraising evidence as for food consumption (see relevant section above), except from the fact that the external validity of these studies will not be appraised as considered not relevant. The critical appraisal tools that will be used are reported in the following tables.

**Table 27. Critical Appraisal Tool for assessing the risk of internal bias (internal validity) of the studies included in the assessment.**

Appraisal question (internal validity)	Rating	Rationale for answering the question
<p>1. Is the handling of the samples for measuring the relevant outcome (endpoint) reliable?</p> <p>(relevant only when the analysis is done in the lab.; not relevant for methods that do not require handling e.g. on-site measurement using hand-held refractometer)</p>	Definitely low RoB	<p>There is a clear reference to a standardized method (e.g. described in relevant SOPs of the lab) for handling the samples applied in the study</p> <p><b>OR</b></p> <p>The method for the handling the samples is well detailed and is considered appropriate (e.g. the samples were kept in cold conditions during the transfer and before the analysis, appropriate labelling system were followed, etc). Ideally, an estimation of the appropriateness of the method is given in the study and is satisfactory (e.g. storage stability)</p>
	Probably low RoB	<p>There is no clear reference for a standardized method for handling the samples, but a description is available and the method applied does not raise concerns</p> <p><b>OR</b></p> <p>The method for handling the samples presents only minor issues in terms of reliability</p>
	Probably high RoB/NR	<p>The method for handling the samples is not described at all (no reference for standardized method, no description, i.e. not reported (NR))</p> <p><b>AND</b></p> <p>there are concerns about the conduct of the study, which indicate that the handling of the samples may not be appropriate</p>
	Definitively high RoB	<p>The methodology for handling the samples is clearly poorly reliable (e.g. there are indications for mix up of the samples, change in sugar content during handling had happened, other artefacts that may alter the accuracy of the outcome are reported)</p>

Appraisal question (internal validity)	Rating	Rationale for answering the question
<p>2. Is the analytical method for measuring the relevant outcome (endpoint) accurate?</p> <p>Note: this is a key appraisal question, i.e. a question that has a higher impact on the overall internal validity of a study for a specific outcome.</p>	Definitely low RoB	<p>There is a clear reference for a standardized method that was applied (e.g. validation of HPLC method, instructions of the used enzymatic kit, standard calibration method for refractometry, spectrometry, titration)</p> <p><b>OR</b></p> <p>The method for measuring the outcome is considered suitable to get an accurate estimation of the outcome in the conditions of the study. Ideally, an estimation of the accuracy of the method is given and it is satisfactory.</p>
	Probably low RoB	<p>There is no clear reference for an applied standardized method, but the method for measuring the outcome does not raise concerns about the accuracy of the measurement.</p> <p><b>OR</b></p> <p>The method for measuring the outcome presents only minor issues in terms of accuracy.</p>
	Probably high RoB/NR	<p>There are concerns that the method may not provide an accurate estimation of the outcome available</p> <p><b>OR</b></p> <p>The method for measuring the outcome is not described (NR)</p>
	Definitively high RoB	The methodology for measuring the outcome is very poor.
<p>3. Is statistical analysis appropriate and consistent with the initial proposal of the study?</p> <p>(applicable only when the statistical results of the study are used. If raw data are provided, the statistical analysis will not be assessed as the data will be analysed separately)</p>	Definitely low RoB	Clear evidence of appropriate statistical analysis
	Probably low RoB	It can be inferred that the statistical analysis is appropriate
	Probably high RoB/NR	<p>It is not possible to assess the appropriateness of the statistical analysis</p> <p><b>OR</b></p> <p>The statistical methods are not reported at all and cannot be inferred</p>



<b>Appraisal question (internal validity)</b>	<b>Rating</b>	<b>Rationale for answering the question</b>
Note: It is expected that in very rare cases, some statistical analysis other than basic statistics (e.g. mean and SD) will be available (without the availability of the raw data) and will have to be appraised	Definitively high RoB	Direct evidence of inappropriate statistical analysis

### 5.3.5.1. Presentation of the results of evidence appraisal, by endpoint

The results of evidence appraisal by endpoint will be presented in tabular format both for individual studies and at the level of the body of evidence (using e.g. an heatmap as the one presented below). In the former case, the rationale for the rating of each appraisal question will also be reported.

**Table 28. Example of heatmap for presenting the results of evidence appraisal across the body of evidence**

Reference metadata	Endpoint(s)	Results	Internal validity		
			Method for handling the samples (if applicable)	Analytical method for measuring the endpoint (key question)	Statistical analysis (if applicable)
			DLRoB	DLRoB	DHRoB
			DHRoB	DHRoB	na
			na	PHRoB	PLRoB

DH: definitively high; PH: probably high; PL: probably low; DL: definitively low; RoB: risk of bias; na: not applicable

### 5.3.6. Data extraction from the included studies and evidence synthesis

Studies may be retained for or discarded from the synthesis depending on the results of critical appraisal. If retained for the synthesis, studies will undergo a structured data extraction based on pre-defined data models and related instructions in DistillerSR®. Before implementation, the data extraction model will be pilot tested on a subset of studies and refined accordingly.

The present analysis attempts to provide answers to a single and rather straightforward assessment question. The expected outcome is expected to be rather homogeneous, although some assumptions may be necessary to convert different ways of expressing sugar content to a common currency (e.g. weight/weight concentration).

It is expected that the degree of aggregation of results will vary across the various publications. For instance, in some studies the results can be reported in an aggregated way (e.g. multiple samples collected over a time period, aggregated in one value), while in some other cases the results can be split by time of sampling. This will be reflected in the data extraction and subsequent analyses

Examples of variables that will be extracted (if reported) from the included studies are: study type; variety of the crop; date and time of the study/sampling as precise as possible (up to hours sampling, if reported); date and time of the flowering period (in order to estimate the age of the flower sampled); sampling method (directly from plant or by using bees, which

kind of bees); number of (sub)samples represented by the value; study location; air humidity;

Considering the nature of the outcome variable of interest, a generalized linear mixed model could be used to estimate sugar content by crop accounting for other factors, such as landscape, environmental conditions, etc. The final choice of the method for synthesising evidence will depend on the granularity and heterogeneity of the studies retrieved.

If all studies are retained for the synthesis and they present different degrees of (internal and external) validity, subgroup analyses may be carried out to explore the impact on the final estimation of the parameter of interest. If subgroup analyses lead to different conclusions, an expert knowledge elicitation might be considered to reach an overall conclusion and characterise the related uncertainties. It is anticipated that depending on the amount, validity and heterogeneity of the available evidence, it may not be possible to conduct quantitative syntheses.

## **5.4. Residue levels in pollen/nectar and TWA**

### **5.4.1. Background**

In 2017, the external scientific report on the "Collection and analysis of pesticide residue data for pollen and nectar" EFSA Supporting publication 2017:EN-1303 (Kyriakopoulou et al., 2017) was published. Under the scope of the project the contractors collected new available information derived from residue trials, evaluated and used these data in order to create a database for pesticide residue levels and residue decline in pollen and nectar, sugar and protein content of nectar and pollen respectively. Additionally, a data analysis of the collected data was performed in order to identify potential correlations between the residue levels and decline in pollen and nectar and between residue levels in pollen and nectar and physicochemical and environmental fate and behaviour properties of the substances. Data submitted and evaluated for the peer-review process under Reg. (EC) 1107/2009) from the year 2010 until 2016 were considered in the project. In total 125 studies were evaluated. Each one of the identified studies was evaluated according to the assessment protocol developed based on the principles of Appendices G and S of the EFSA's bee guidance document (EFSA, 2013). RUD values were calculated considering the measured initial residue values in each matrix and the respective application rates or seed dressing rates. For studies and matrices where the residue dissipation was followed by sampling in a sufficient number of time points after pesticide application, DT50 and DT90 values for each matrix were calculated as well.

Data from open literature were not considered in this project. This is because in order to derive a meaningful RUD value and estimate the dissipation rate, some specific study conditions have to be fulfilled. The residue levels have to be linked to a certain pesticide application that happened in a certain field and in certain time. Also, in order to use the data in a common data base, a certain level of comparability is needed as regards to the study design and reporting (field and site selection, agricultural practices, sampling methods and regime, samples handling and analysis, reporting of the used methods and environmental conditions, reporting of the results and the data analysis etc.). Data submitted under Reg. (EC) 1107/2009) usually fulfils these requirements, while open literature studies are usually designed for monitoring purposes (i.e. informs about residue levels in bee relevant matrixes, but no RUD values can be derived).

It should be noted that, at the time of the compilation of this protocol, the data collection for this review had already been finalized while the data analysis is still to be done.

#### 5.4.2. Aim of the review

The aim of this review, as agreed by the working group, is to amend the existing database for the initial residue values (derivation of additional RUD values) and on the dissipation rate of the different pesticide molecules from pollen and nectar that was developed for the EFSA Supporting publication 2017:EN-1303 (Kyriakopoulou et al., 2017). The amended database will be used for the exposure estimation in the tier 1 model. As agreed by the working group, other aspects that were included in the original projects (e.g. sugar content of the nectar, protein content of the pollen) will not be amended due to the very limited data that was available or was generated in the dossier studies in the period between 2017-2019). Also, for the same reason, it was decided that the data base only for spray applications will be amended.

#### 5.4.3. Question formulation

The question that will be answered through the systematic review is the same as in section 1 of the EFSA Supporting publication 2017:EN-1303 (Kyriakopoulou et al., 2017).

#### 5.4.4. Eligibility criteria and evidence appraisal

The assessment protocol as described in EFSA Supporting publication 2017:EN-1303 (see section 2 of the external report) was used without modifications for the evaluation of the studies.

#### 5.4.5. Search strategy, study selection and data extraction

All studies measuring residues in relevant matrices available in active substance dossiers submitted between 2017 and 2019 were gathered by EFSA. Exclusion of those studies that were already evaluated in the context of the original project resulted in a list of 28 new studies, performed with the following active substances: acetamiprid, tebuconazole, alpha-cypermethrin, dimethoate, isoflucypram, pyrimethanil, dithianon, spinosad, sulfoxaflor and thiacloprid (for more details see table 29). The database was updated with the results of the new studies.

**Table 29. Overview of the studies added to the residue data base**

Study on active substance	Number of trials	Type of study	DT50 calculation possible?
Acetamiprid 1	1	Tunnel	no
Acetamiprid 3	2	Field	no
Acetamiprid 4	1	Field	no
Tebuconazole	1	Field	no
Alpha-cypermethrin 1	1	Field	no
Alpha-cypermethrin 2	1	Field	no
Alpha-cypermethrin 3 a	1	Field	no
Alpha-cypermethrin 3 b	1	Field	no
Alpha-cypermethrin 3 c	1	Field	no
Alpha-cypermethrin 3 d	1	Field	no
Alpha-cypermethrin 3 e	1	Field	no
Dimethoate 1	6	Tunnel	no
Dimethoate 2	6	Tunnel	no
Isoflucypram 1	3	Tunnel	no
Isoflucypram 2	3	Tunnel	no

Pyrimethanil 1	4	Tunnel	no
Pyrimethanil 2	5	Tunnel	yes
Pyrimethanil 3	1	Field	no
Dithianon	5	Tunnel	yes
Spinosad 1	1	Tunnel	no
Sulfoxaflor 1	4	Tunnel	no
Sulfoxaflor 2	4	Tunnel	yes
Sulfoxaflor 3	4	Tunnel	yes
Sulfoxaflor 4	4	Tunnel	no
Sulfoxaflor 5	1	Tunnel	no
Sulfoxaflor 6	1	Field	no
Sulfoxaflor 7	4	Tunnel	no
Sulfoxaflor 8	1	Glasshouse	no
Thiacloprid 1	1	Field	no
Thiacloprid 2	2	Field	no

#### 5.4.6. Analysis of the amended database

The samples collected immediately after application will be used to build a dataset of initial residue levels for all the relevant matrices. The data will be analysed with a linear mixed model, in order to account for all the relevant factors (active substance, crop, etc.). The results of the analysis will be used to generate a predictive distribution of initial RUD values. Different predictive distributions will be generated for nectar and pollen and for each application method (upward/downward foliar application, seed treatment and granule application) for which enough data are available.

The results for residue decline in EFSA Supporting publication 2017:EN-1303 (Kyriakopoulou et al., 2017) will be updated by including DT50/DT90 values calculated from the new studies.

#### 5.5. Methods to update the exposure related parameters: $f_{dep}$ and $E_f$

The exposure related parameters used in the EFSA, 2013 which are applied to the weed scenario to reflect the fractions of the pesticide mass that deposits to the weeds (i.e.  $f_{dep}$  for the contact exposure assessment and the  $E_f$  parameter for the dietary exposure assessment) will be updated in order to be aligned with the crop interception values used in other exposure assessments. The revised crop interception values proposed by the FOCUS surface water Repair WG will be used (Table 30).

**Table 30.- Crop interception (%) depending on the growth stage (adapted from the Scientific report of EFSA on the “repair action” of the FOCUS surface water scenarios, in preparation)**

BBCH stage	BBCH stage										
	Germination / sprouting	Leaf development			Formation of side shoots	Stem elongation / rosette growth	Vegetative plant parts	Inflorescence emergence	Flowering	Development of fruit	Ripening
BBCH stage	00-09	10-13	14-19	20-29	30-39	40-49	50-59	60-69	70-79	80-89	90-99
Cereals, spring	0	0	0	20	80	90	90	90	80	80	80
Cereals, winter	0	0	0	20	80	90	90	90	80	80	80
Citrus <sup>(a)</sup>	80	80	80	-	80	-	80	80	80	80	80
Cotton	0	30	30	60	60	-	75	75	75	75	90
Field beans	0	25	25	40	-	-	70	70	70	70	80
Grass/alfalfa <sup>(a,b)</sup>	90	90	90	90	90	90	90	90	90	90	90

Grass below the crop canopy in pome/stone fruits and vines <sup>(c)</sup>	45	45	45	45	45	45	45	45	45	45	45
Hops <sup>(d)</sup>	0	20	20	50	50	-	60	60	70	70	70
Legumes <sup>(f)</sup>	0	35	35	-	55	-	85	85	85	85	85
Maize	0	25	25	-	50	-	75	75	75	75	90
Oil seed rape, spring	0	40	40	80	80	-	80	80	80	80	90
Oil seed rape, winter	0	40	40	80	80	-	80	80	80	80	90
Olives <sup>(a,g)</sup>	70	70	70	-	70	-	70	70	70	70	70
Pome/stone fruits	50	60	60	-	60	-	60	60	65	65	(#) <sup>(e)</sup>
Potatoes	0	15	15	60	60	-	85	85	85	85	50
Soybeans	0	35	35	55	-	85	85	85	85	85	65
Sugar beets <sup>(h)</sup>	0	20	20	-	70	90	90	90	90	90	90
Sunflower	0	20	20	-	50	-	75	75	75	75	90
Tobacco	0	50	50	70	-	-	90	90	90	90	90
Vegetables, bulb <sup>(h,i)</sup>	0	10	10	-	-	40	40	40	40	40	60
Vegetables, fruiting <sup>(j)</sup>	0	50	50	70	-	-	80	80	80	80	50
Vegetables, leafy <sup>(h,k)</sup>	0	25	25	-	-	70	70	70	70	70	90
Vegetables, root <sup>(h,l)</sup>	0	25	25	-	-	80	80	80	80	80	80
Vines	40	50	60	-	-	-	60	60	75	75	(#) <sup>(e)</sup>

Non-existing crop stages according to the BBCH Compendium are represented by a dash (-)

- (a): Evergreen all year round, constant crop interception not related to BBCH stage
- (b): Considered to represent established turf all year round
- (c): In line with revised runoff parameterisation for tall permanent crops in PRZM; notice that there is no soil cover (bare soil, no crop interception) below the crop canopy in citrus, hops and olives
- (d): Adopted from Olesen and Jensen (2013); according to this reference crop interception at BBCH 92 and 97 is 70 % and 0 %, respectively (no other BBCH stage exists in hops for BBCH 90-99)
- (e): No crop interception given in EFSA (2014) for this BBCH stage; default crop interception set to zero
- (f): Same as peas in FOCUS groundwater
- (g): Same as in FOCUS surface water Step 2
- (h): Harvested at BBCH 50; default crop interception set to zero for BBCH 50 and onwards
- (i): Same as onions in FOCUS groundwater
- (j): Same as tomatoes in FOCUS groundwater
- (k): Same as cabbage in FOCUS groundwater
- (l): Same as carrots in FOCUS groundwater

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## **Glossary [and/or] Abbreviations**

**Glossary:** an alphabetical list of words relating to a specific subject, text, or dialect, with explanations; a brief dictionary.

**Abbreviation:** a shortened form of a word or phrase (such as Mr., Prof.). It also includes acronyms (a group of initial letters used as an abbreviation for a name or expression, each letter being pronounced separately – such as DVD, FDA – or as a single word – such as EFSA, NATO).

## Appendix A – Sensitivity analysis

### A.1. Introduction

The sensitivity analysis considered the following equation (adapted from equations 3 and 4):

$$SV = PFF TWA \frac{(LDFp CONCp CONSp) + \left( LDFn CONCn \frac{CONSsugar}{sugar \text{ in } n} \right)}{1000} \quad [\text{eq. A1}]$$

Shortcut values for Tier 1 (henceforth,  $SV_{90}$  values) are calculated as the 90<sup>th</sup> percentile of the distribution of SV, given a plausible range of variability for the input parameters.

The aim of the sensitivity analysis was to identify, among the nine input parameters in eq. A1, the main drivers of the value of  $SV_{90}$ . The preliminary steps consisted in: identifying/estimating appropriate probability distributions for all the input parameters; using those to generate a distribution of SV values from Eq. A1; calculating the  $SV_{90}$ . (“baseline” value) from that distribution. The goal of the subsequent sensitivity analysis was to investigate the contribution of each input parameter (and distribution thereof) to the  $SV_{90}$  value, and to identify the most influential parameters. The results of this sensitivity analysis were considered by the WG for the prioritisation of parameters.

### A.2. Methods

#### A.2.1. Input parameters

A requirement of this analysis is that the variation of each input parameter be described by a probability distribution. For two parameters, probability distributions estimated from data were already available and were used. For all the other input parameters this was not the case, as only ranges of variation were available. The ranges had to be converted to probability distributions, which implied unavoidable extra assumptions as explained below. In order to counter any bias related to those extra assumptions, it was decided to consider two alternative distribution scenarios, using either normal distributions or uniform distributions for all the parameters. The sensitivity analysis was run for both scenarios.

##### A.2.1.1. Selection of ranges

For  $CONCn$  and  $CONCp$ , estimated distributions were already available (see below) and there was no need to consider ranges. Ranges of variation for all the other input parameters were either extracted from the 2013 Guidance, or calculated from existing data, or based on expert knowledge.

Specifically, for sugar and pollen consumption ( $CONSsugar$  and  $CONSp$ ), ranges for the relevant bee groups and life stages were provided in the 2013 Guidance (Appendix J, Table J1). The range of variability for TWA was extracted from Kyriakopoulou et al. (2017), where  $DT_{50}$  values calculated for several active substances covered a range of 0.5-4.5 days. Based on this interval, the ranges for TWA were calculated as 0.07-0.51 for adult bees (based on a 10-day exposure) and 0.14-0.70 for larvae (based on a 5-day exposure). Based on available

knowledge, the range for sugar content of nectar was 10%-65%; the range for the landscape dilution factor (LDFp and LDFn for pollen and nectar, respectively) was set to 0.5-1; and the range for the pre-flowering factor (PFF) was set to 0-1.

Parameter	Range	Probability distributions used in the analysis
CONSugar, CONSp	Ranges from Appendix J, Table J1 of the 2013 Guidance	Derived from range (when available): (1) truncated normal (2) uniform
CONCn, CONCp	Not applicable	Lognormal distributions as in Appendix J, Table J2 of the 2013 Guidance (parameters for downward spraying)
Sugar in nectar	10-65%	Derived from range: (1) truncated normal (2) uniform
PFF	0-1	Derived from range: (1) truncated normal (2) uniform
LDFp, LDFn	0.5-1	Derived from range: (1) truncated normal (2) uniform
TWA	0.07-0.51 (adult bees) 0.14-0.70 (larvae) (calculated from a range for DT50 of 0.5-4.5 days)	Derived from range: (1) truncated normal (2) uniform

#### A.2.1.2. Choice of probability distributions

A probability distribution for CONCn and CONCp (residue concentration on nectar and pollen) was already available in the 2013 Guidance, where lognormal distributions were estimated from available data. The parameters used in this analysis are those for downward spraying in Appendix J, Table J2.

For the other parameters, variability was initially expressed as a range (min-max). For the purpose of the analysis, it was necessary to make hypotheses on the underlying probability distribution. Two alternative options were considered:

- The parameter is normally distributed: the mean coincides with the midpoint of the initial range and the .5/99.5 percentile with the lower/upper limit, so that 1% of the values fall outside the range. This is the same assumption used for some of the parameters in the 2013 Guidance (Appendix J) for the calculation of Tier 1 values. The distributions are appropriately truncated to avoid non-allowed (e.g. negative) values.
- The parameter is uniformly distributed between the lower and upper limit of the initial range. With respect to the previous one, this choice implies weaker assumptions (all the values within the range are equally likely) and it is considered a good alternative as it generates distributions with a larger variance.

In the sensitivity analysis, the choice between normal and uniform distribution was taken for all the parameters together: hence, the analysis was done twice, once for each of the **two following scenarios**:

**Normal:** normal distribution for all parameters; lognormal distribution for CONCN and CONCP

**Uniform:** uniform distribution for all parameters; lognormal distribution for CONCN and CONCP

The main motivation for considering two scenarios was that the outcome of a sensitivity analysis might be affected by (and confounded with) the specific choice of distribution. Comparing results between the two scenarios allows identifying common trends. The lognormal distributions for CONCN and CONCP were not changed between the two scenarios; as no strong evidence was found against the current choice of distribution, these two parameters were not tested for alternative distributions.

## A.2.2. Sensitivity analysis

### A.2.2.1. Generating the distribution of SV values

A Monte Carlo simulation method was used to generate the distribution of SV values. The basic step consists in drawing a value for each of the nine parameters from the probability distributions estimated in A.2.1; this set of values is used to calculate a value of SV from equation A1. This step is repeated many times to obtain a large sample (at least  $10^4$ ) of SVs. This is considered the 'baseline' distribution for SV; the 90<sup>th</sup> percentile of this distribution is the baseline value of  $SV_{90}$ .

### A.2.2.2. Identifying the contribution of each parameter

The baseline  $SV_{90}$  value is affected by the variability of the baseline distribution for SV, which in turn is the combined effect of the variability of all the input parameters. The aim of the analysis is to identify, among the parameters, the most relevant sources of variation – i.e. the main drivers in determining the value of  $SV_{90}$ .

The following is an outline of the method (see the calculation of first-order sensitivity indices in Saltelli et al., 2004 for a similar framework). Let X be one of the parameters of the model. The aim is to understand how much the variability of X contributes to the variability of SV (hence, to the value of  $SV_{90}$ ). A way to achieve this is to remove the variability for X and check what changes: set X to a plausible constant value (e.g. the median), generate a new distribution of SVs keeping X fixed and calculate the 90<sup>th</sup> percentile. In general, the variability of the new, fixed-parameter SV distribution should be reduced; as a consequence,  $SV_{90}$  should be decreased with respect to the baseline. A strong decrease would indicate that the variability of X has a strong impact on  $SV_{90}$ ; if the decrease is negligible, the variability of X can be considered uninfluential. In the present analysis, this idea was implemented (item (1.b) in the outline in Section A.2.2.3) following a slightly more complex averaging procedure (details are omitted here).

### A.2.2.3. General overview of the sensitivity analysis

An outline of the analysis is provided here in the form of a series of instructions.

For each **combination bee group/life stage** (e.g. “honey bee larva”) do the following:

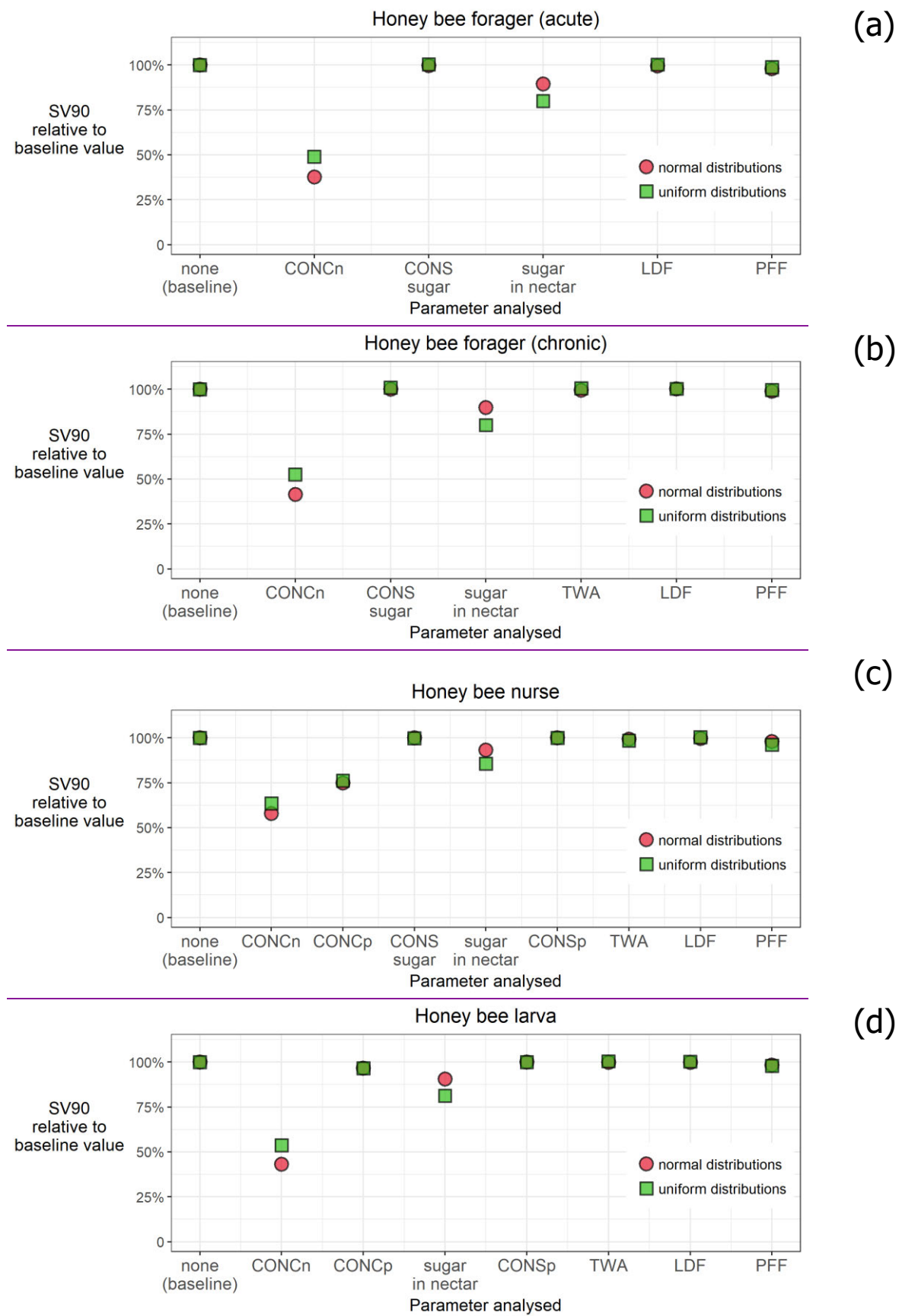
1. For each distribution scenario, **Normal** or **Uniform**
  - (1.a) Generate the baseline distribution of SVs (Section A.2.2.1) using the parameter distributions specific to “honey bee larva” (Table A.1) and to the distribution scenario (Section A.2.1.2).
  - (1.b) For each parameter (X) do the following:  
Calculate a new, fixed-parameter  $SV_{90}$  value by removing the variability of X (see Section A.2.2.2). Calculate the ratio of the fixed-parameter  $SV_{90}$  value to the baseline value. (The new value will be in general lower than the baseline; the lower it is, the stronger the influence of X.)
  - (1.c) When all the parameters have been analysed in (1.b), plot all the results in a single graph: fixed-parameter  $SV_{90}$  values (expressed as % of the baseline  $SV_{90}$  value) for each parameter. Compare results among parameters to identify the most relevant.
2. When both **Normal** and **Uniform** scenarios have been investigated, plot all the results in a single graph for comparison (i.e. merge the two graphs generated in (1.c)). This is the graph presented in this Appendix. Trends in the results common to both scenarios should be considered robust with respect to the choice of distribution.

### A.3. Results

The results of the sensitivity analysis are shown in Figures A1-A3.

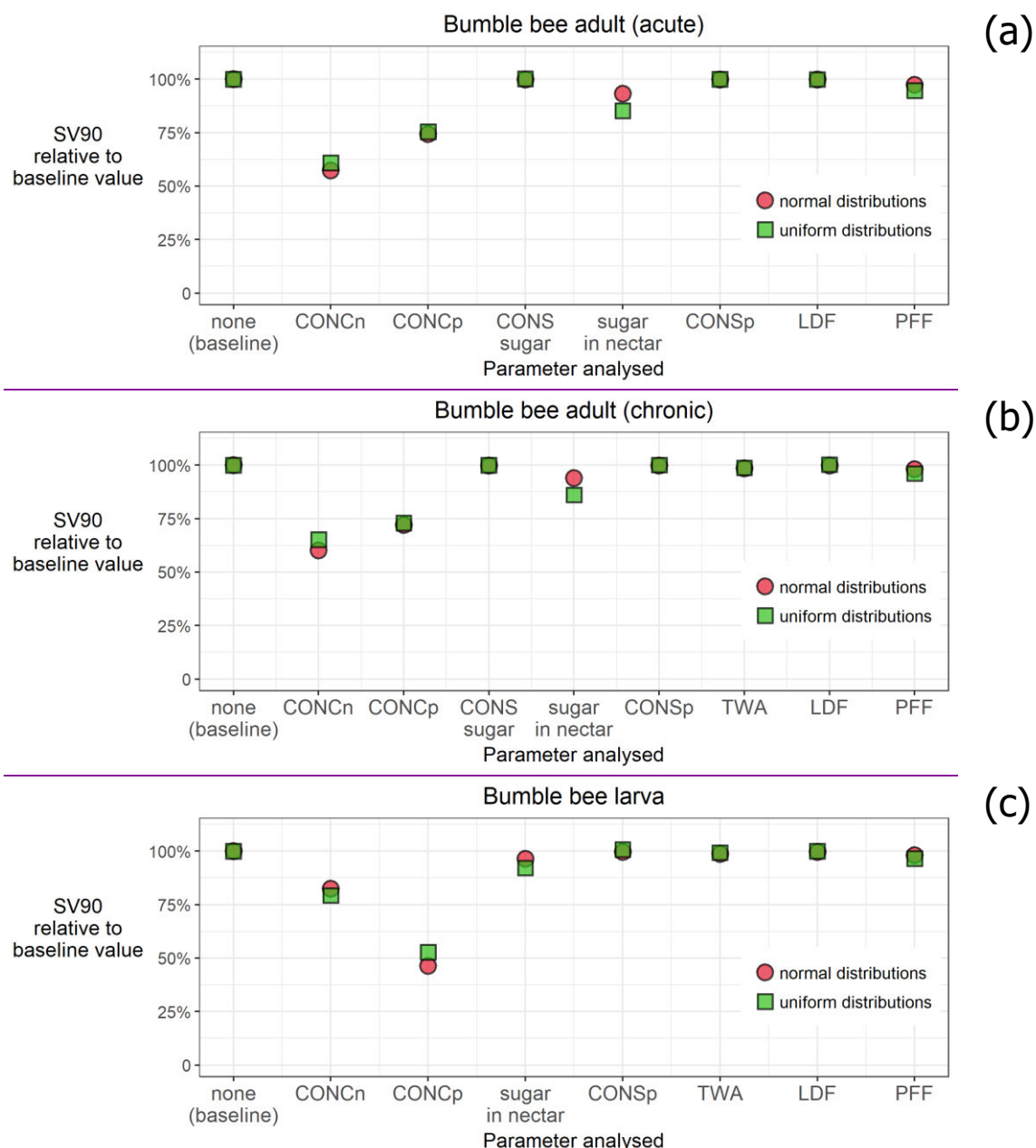
The common trend emerging from the results is that CONCN and CONCP have the strongest influence, with a relative decrease of  $SV_{90}$  up to 70% for CONCP and solitary bee larvae. Although differences exist between the decrease in the **Normal** and the **Uniform** scenarios, the results are always consistent. Sugar in nectar also has a (weaker) influence on  $SV_{90}$ . The other parameters do not rank as influential in the current analysis.





**Figure A1.** Results of the sensitivity analysis for honey bees. **(a)** Results for foragers (acute exposure). For each parameter, fixed-parameter SV<sub>90</sub> values were estimated for two different distribution scenarios, “normal” (red circles) and “uniform” (green squares) and are

expressed here as % of the baseline  $SV_{90}$  for ease of comparison. **(b-d)** Results for all appropriate combinations of different life stages (larvae and adults), different castes for adults (workers and foragers) and different exposure scenarios for foragers (chronic). The conventions in each graph are the same as in (a). The strong decrease from the 100% value  $CONC_n/CONC_p$  indicates that the parameters have a strong influence on  $SV_{90}$ ; a smaller decrease is also consistently observed for sugar in nectar.



**Figure A2.** Results of the sensitivity analysis for bumble bees for appropriate combinations of different life stages (larvae and adults) and different exposure scenarios for adults (acute and chronic). The conventions in each graph are the same as in Figure (A1).



**Figure A3.** Results of the sensitivity analysis for solitary bee larvae and adults. The conventions in each graph are the same as in Figure (A1).

## Appendix B – Method for the EKE on crop attractiveness (active pollen collection)

### B.1. Methods for performing Expert Knowledge Elicitations

One EKE session is planned in summer 2020 in EFSA's headquarter in Parma (Italy). All results will be published in connection with the revision of the Guidance Document.

#### B.1.1. Timeline

**Table 1:** Timeline for the EKE session

Step	Date
Draft protocol for review	End March 2020
Long list of possible experts	End April 2020
Final protocol	Mid May 2020
Draft evidence dossier for review	End May 2020
Invitation of Experts	End May 2020
EKE session	Summer 2020
Internal result report for review	August 2020

#### B.1.2. EKE Questions

Following definitions are proposed:

**Table 2:** Definitions of terminology used in the EKE question

Term	Definition
Honey bees	European Honey bees (single species: <i>Apis mellifera</i> , with several sub-species). These are eusocial bees living in generally large, perennial colonies with a single egg-laying queen. The focus for this EKE is on forager bees. Foragers collect the food items, pollen and nectar (occasionally extrafloral nectarine, honey dew) from crops and plants available in the environment. They bring these items back to the hive and give them to the in-hive bees. They collect also other items that may be necessary for the colony like resin or water. Honey bees have sophisticated communicational channels; with the waggle dance the scouts and foragers communicate the location and the quality of the foraging site to the other foragers.
Bumble bees	Bumble bees (species belonging to a single genus: <i>Bombus spp.</i> ). Most bumble bee species are eusocial, with bees living in annual colonies with a single founding queen. Similarly to honey bees, some workers forage for food. The communication between bumble bees is more trivial compared to honey bees; it is not known for example whether bumble bees are able to communicate the foraging site to each other.
Solitary bees	Solitary bees (taxonomically non-homogeneous group). This group includes the majority of bee species, characterised by solitary behaviour. The high diversity of this group is not only limited to taxonomy, but it is also reflected in many ecological traits: nesting, feeding, and reproduction strategy can be very different. As for the other bees, adult solitary bees feed on pollen and nectar
Northern zone	One of the regulatory zones in which the EU is divided for the authorisation of plant protection products, as defined in Annex I of Reg. 1107/2009. The following Member States belong to this zone: Denmark, Estonia, Latvia, Lithuania, Finland, Sweden.
Central zone	One of the regulatory zones in which the EU is divided for the authorisation of plant protection products, as defined in Annex I of Reg. 1107/2009. The following Member States belong to this zone: Belgium, Czech Republic, Germany, Ireland, Luxembourg, Hungary, Netherlands, Austria, Poland, Romania, Slovenia, Slovakia, United Kingdom. It is noted that after 31/01/2020, the United Kingdom is no longer formally part of the EU. However, for the sake of this exercise, the country is tentatively still considered as part of the Central zone

Southern zone	One of the regulatory zones in which the EU is divided for the authorisation of plant protection products, as defined in Annex I of Reg. 1107/2009. The following Member States belong to this zone: Bulgaria, Greece, Spain, France, Italy, Cyprus, Malta, Portugal.
Crop production	Crop grown with the main aim of producing food, feed, fibre, or fuel. They may be harvested before flowering when the focus of the production (e.g. edible part) is developed before BBCH 60 (e.g. carrots).
Seed production	Crop grown with the main aim of producing seeds for generation of future crops. They are always harvested after flowering irrespectively of the development time of the edible part.

The WG proposes alternative EKE questions. Both will be left in the protocol for the sake of receiving comments during the public consultation. On the basis of the feedback received during this phase and further discussions, the WG will make the final decision.

**Table 3:** Proposed framing of the EKE question

Topic	Description	
Parameters	<b>Proportion of nests with active pollen collection from the target crop</b>	<b>Average pollen collection from the target crop</b>
Strata	Target crop (Genus, species/cultivar, agricultural practice) Bees (Honey bees, bumble bees, solitary bees) Regions (Northern, Central, Southern European regulatory zones)	
Definition	Pollen is actively collected, if the proportion of pollen of the [target crop] collected by one [honeybee colony / bumble bee nest / solitary bee] is more than 5%.	The average proportion is calculated as the part of pollen of the [target crop] in relation to the total of pollen collected by all [honeybee colonies / bumble bee nests / solitary bees].
Question	What is the proportion of [honeybee colonies / bumble bee nests / solitary bees] that actively collect pollen of the [target crop] in the [Northern / Central / Southern] regulatory European zone?	What is the average proportion of pollen of the [target crop] collected by [honeybee colonies / bumble bee nests / solitary bees] in the [Northern / Central / Southern] regulatory European zone?
Unit	[out of 1000] nests	[%] pollen
Operationalisation	Imagine 1000 [honeybee colonies / bumble bee nests / solitary bees] placed next to the [target crop] production field - during the life stage, when pollen is produced (peak flowering bloom: BBHC Stage 65-67) - in the [Northern / Central / Southern] regulatory European zone. The bees are allowed to forage freely from the field or the surrounding area.  The number of [honeybee colonies / bumble bee nests / solitary bees] (out of the 1000) with more than 5% of the gathered pollen originating from the [target crop] is the answer.	Imagine 1000 [honeybee colonies / bumble bee nests / solitary bees] placed next to the [target crop] production field - during the life stage, when pollen is produced (peak flowering bloom: BBHC Stage 65-67) - in the [Northern / Central / Southern] regulatory European zone. The bees are allowed to forage freely from the field or the surrounding area.  All pollen entering the hives/nests is examined. The proportion of pollen originate from the [target crop] is the answer
Threshold	Final conclusion on "bee attractivity" is made using the threshold of "more than 10%" of [honeybee colonies / bumble bee nests / solitary bees] are exposed	The question allows a ranking between crops, but a clear threshold is not defined.
Remarks	The limit 5% is set by the WG to distinguish "active collection" from "random contamination" The approach does not consider the total volume of collected pollen, as only information, if "active collection" is given, is used.	Please note that an average below 5% of the collected pollen does not imply that less than 10% of the [honeybee colonies / bumble bee nests / solitary bees] are exposed. Information on the total pollen of collection is needed, this means that [hives/nests/bees] with higher volumes count more to the average.

### B.1.3. EKE Experts

**Table 4:** Constitution of the proposed expert panel

Expert profile	Description
Bee expert North Europe	Researcher specialized in bee behaviour and overview on North European cropping practices / landscapes
Bee expert Central Europe	Researcher specialized in bee behaviour and overview on Central European cropping practices / landscapes
Bee expert Southern Europe	Researcher specialized in bee behaviour and overview on Southern European cropping practices / landscapes
Crop expert for "Wind pollinators"	Researcher specialized on wind pollinating crops, esp. cereals
Crop experts for other specific plants	

**Table 5:** Expertise matrix

	Knowledge		Role			Region			Matrices	
	general	specific	academic	governmental	practitioner	Northern Europe	Central Europe	Southern Europe	bees	crops
1 <sup>st</sup> expert		X	X			X			X	
2 <sup>nd</sup> expert		X	X				X		X	
3 <sup>rd</sup> expert		X	X					X	X	
4 <sup>th</sup> expert	X		X						(x)	X
5 <sup>th</sup> expert	X		X						(x)	X
6 <sup>th</sup> expert	X		X						(x)	X

(x) indicates a basic knowledge (i.e. broad knowledge of bee biology and ecology)

#### B.1.4. EKE Methodology

EFSA's Expert Knowledge Elicitation is a structured approach to retrieve expert judgements from a group of experts, especially selected for the question of interest. Questions are usually asking for a quantitative parameter, the expected answer includes the consensus of the group of experts and a description of their remaining uncertainties about their judgements.

The panel proposes to use an adapted version of the Sheffield methodology for the elicitation session. This methodology uses a face-to-face meeting of the experts with behavioural aggregation to find a consensus result. The session will be led by a facilitator.

- Step 0: Introduction to the context of the elicitation, task of the session, and methodology of the elicitation. This includes a training of the experts on probabilistic judgements.
- Step 1: Clarification on the elicitation question and underlying definitions
- Step 2: Review and discussion of the evidence dossier to answer the question
- Step 3: Identification of limitation of the evidence to answer the question
- Step 4: Construction of a high and low value scenario for the answer
- Step 5a: Individual judgements on the credibility range of the answer (upper and lower limit of the answer)
- Step 5b: Disclosure of the individual answers to the group
- Step 5c: Behavioural aggregation to find a consensus on the credibility range, incl. discussion on the importance of uncertainties defining the high and low value scenario. Summary reasoning of the result.
- Step 6a: Individual judgements on a fair estimate (median: equally over- or under estimating), and the precision (interquartile range) of the judgement
- Step 6b: Feedback of the individual judgements / uncertainty distributions to the group
- Step 6c: Behavioural aggregation to find a consensus on the median and interquartile range of the judgements. Summary reasoning on the result.
- Step 7: Fitting of the final distribution, review of the result and approval of the group
- Step 8: Review of the reasoning and result report of the EKE session



### B.1.5. Draft Agenda

The duration will depend on the number and diversity of remaining crops for elicitation

**Table 6:** Proposed agenda of the EKE session (3 days)

Time	Topic
<b>1<sup>st</sup> day</b>	
AM	Introduction to task and methodology, incl. training of the experts
PM	Review of retrieved evidence
<b>2<sup>nd</sup> day</b>	
AM	EKE
PM	
<b>3<sup>rd</sup> day</b>	
AM	Review of the result report

## B.2. Methods for result reporting

### B.2.1. Fitting the continuous distribution

For each elicited parameter an uncertainty distribution should be derived. It is proposed to use a fitted continuous distribution function to allow the interpolation of percentiles, which were not directly elicited.

For each parameter and the final result of the fitting, the density function and the descending distribution function, as well as the theoretical formula will be documented.

A table of selected percentiles will summarize the elicited parameters, the fair estimate (median: equally over- or under estimating), and different levels of remaining uncertainties, e.g. the 90% uncertainty range.

### B.2.2. Reporting of uncertainties.

Appropriate graphs (e.g. CDFs) are used to compare the results of different crops and show the relation of the final selection from the certainty level.

## Appendix C – Search strings on food consumption

Three different search strategies have been designed to cover the 4 sub-questions related to food consumption, all of them share the same search strings for population. The search strategies by database have been included in the same table to simplify the reporting. The *comments* column links the number of question/s to each search strategy.

### Biosis

Set	Query	Comments
# 15	#14 AND #4 Indexes=BCI Timespan=All years	Q4
# 14	#13 OR #12 Indexes=BCI Timespan=All years	
# 13	TI=((behavior* OR behaviour* OR distance* OR time* OR length* OR duration* OR performance* OR abilit*) AND (fly* OR flight* OR trip OR trips)) OR ((forag* OR collect* OR search*) AND ( fly* OR flight* OR trip OR trips) ) OR (activit* AND (forag* OR monitor* OR track*)) Indexes=BCI Timespan=All years	
# 12	TS=((behavior* OR behaviour* OR distance* OR time* OR length* OR duration* OR performance* OR abilit*) NEAR/5 (fly* OR flight* OR trip OR trips)) OR ((forag* OR collect* OR search*) NEAR/5 ( fly* OR flight* OR trip OR trips) ) OR (activit* NEAR/5 (forag* OR monitor* OR track*)) Indexes=BCI Timespan=All years	
# 11	#10 AND #4 Indexes=BCI Timespan=All years	Q3
# 10	TS=(energ* AND (collection* OR consumption OR cost OR costs OR demand* OR efficienc* OR forag* OR intak* OR need* OR performance )) Indexes=BCI Timespan=All years	
# 9	#8 OR #6 Indexes=BCI Timespan=All years	Q1 – Q2
# 8	#7 AND #4 Indexes=BCI Timespan=All years	
# 7	TI=((food* OR nectar* OR pollen* OR sugar OR carbohydrate*) AND ("consume" OR "consumed" OR "consumes" OR consumption* OR intak* OR feed* OR diet OR diets OR dietar*)) Indexes=BCI Timespan=All years	
# 6	#5 AND #4 Indexes=BCI Timespan=All years	
# 5	TS=((food* OR nectar* OR pollen* OR sugar OR carbohydrate*) NEAR/5 ("consume" OR "consumed" OR "consumes" OR consumption* OR intak* OR feed* OR diet OR diets OR dietar*)) Indexes=BCI Timespan=All years	
# 4	#3 OR #2 OR #1 Indexes=BCI Timespan=All years	
# 3	TS=((Europe* OR Euroregion* OR "Euro region*" OR Austria* OR Belgium OR Belgian OR Bulgaria OR Bulgarian OR Croat* OR Cyprus OR Cypriot* OR Czech* OR Denmark* OR Danish OR Estonia* OR Finland OR finnish* OR France OR French* OR German* OR Greece OR "hellenic republic" OR Greek* OR Hungar* OR Ireland OR Irish OR Ital* OR Latvia OR Lithuania* OR Luxembourg* OR Malta OR maltese* OR Netherlands* OR dutch* OR Poland* OR Polish* OR Portug* OR Romania* OR Slovak* OR Slovenia* OR Spain OR Spanish* OR Swed* OR "United Kingdom" OR Britain OR British OR England* OR English* OR Scotland* OR Scottish OR Wales OR Welsh OR Liechtenstein* OR Iceland* OR Norway* OR Norwegian* OR Switzerland* OR Swiss* OR Albanian* OR Macedonia* OR Montenegro* OR Serbia* OR Bosnia* OR Kosov* OR Benelux OR Czechoslovakia* OR Scandinav* OR Yugoslavia* OR Balkan* OR "Baltic countr*" OR "Baltic state*" OR "Iberian peninsula" OR Iberia OR "Mediterranean countr*" OR "Mediterranean state*" OR "Mediterranean region*" OR "Nordic countr*" OR "Nordic state*") AND (bee OR bees)) Indexes=BCI Timespan=All years	
Q# 2	TS=("managed bee" OR "managed bees" OR "Apis mellifera" OR "Apis mellifica" OR "a mellifera" OR "a mellifica" OR Honeybee* OR "honey bee*" OR Bombus OR Bumblebee* OR "Bumble bee*" OR "B. alpinus " OR "B. argillaceus " OR "B. armeniacus " OR "B. balteatus " OR "B. barbutellus " OR "B. bohemicus " OR "B.	

	<p>brodmannicus " OR "B. campestris" OR "B. cingulatus " OR "B. confusus " OR "B. consobrinus" OR "B. cryptarum" OR "B. cullumanus " OR "B. deuteronymus" OR "B. distinguendus" OR "B. flavidus " OR "B. fragrans " OR "B. gerstaeckeri " OR "B. glacialis " OR "B. haematurus " OR "B. hortorum " OR "B. humilis " OR "B. hyperboreus " OR "B. hypnorum " OR "B. inexpectatus " OR "B. jonellus" OR "B. laesus" OR "B. lapidarius" OR "B. lapponicus" OR "B. lucorum" OR "B. magnus" OR "B. mendax" OR "B. mesomelas" OR "B. mlokosievitzii" OR "B. mocsaryi" OR "B. modestus" OR "B. monticola" OR "B. mucidus" OR "B. muscorum" OR "B. niveatus" OR "B. norvegicus" OR "B. pascuorum" OR "B. patagiatus" OR "B. perezi" OR "B. pereziellus" OR "B. polaris" OR "B. pomorum" OR "B. pratorum" OR "B. pyrenaeus" OR "B. quadricolor" OR "B. reinigiellus" OR "B. ruderarius" OR "B. ruderatus" OR "B. rupestris" OR "B. saltuarius" OR "B. schrencki" OR "B. semenoviellus" OR "B. sichelii" OR "B. soroeensis" OR "B. sporadicus" OR "B. subterraneus" OR "B. sylvarum" OR "B. sylvestris" OR "B. terrestris" OR "B. vestalis" OR "B. veteranus" OR "B. wurflenii" OR "B. zonatus")</p> <p>Indexes=BCI Timespan=All years</p>	
# 1	<p>TS=((solitary NEAR/3 (bee OR bees)) OR Afranthidium OR Aglaoapis OR Amegilla OR Ammobates OR Ammobatoides OR Ancylo OR Andrena OR ANDRENIIDAE OR ANDRENINAE OR Anthidiellum OR Anthidium OR Anthophora OR APIDAE OR APINAE or Biastes OR Camptopoeum OR Ceratina OR Ceylalictus OR Chelostoma OR Chiasmognathus OR Clavipanurgus OR Coelioxys OR Colletes OR COLLETIDAE OR COLLETINAE OR Cubitalia OR Dasypoda OR DASYPODAINAE OR Dioxys OR Dufourea OR Ensliniana OR Eoanthidium OR Epeoloides OR Epeolus OR Eucera OR Flavipanurgus OR Habropoda OR Haetosmia OR HALICTIDAE OR HALICTINAE OR Halictus OR Heriades OR Hofferia OR Hoplitis OR HYLAENIAE OR Hylaeus OR Icteranthidium OR Lasioglossum OR Lithurgus OR Macropis OR Megachile OR MEGACHILIDAE OR MEGACHILINAE OR Melecta OR Melitta OR MELITTIDAE OR MELITTINAE OR Melitturga OR Metadioxys OR (Nomada AND (bee OR bees)) OR NOMADINAE OR NOMIINAE OR Nomiapis OR NOMIOIDINAE OR Nomioides OR Osmia OR Panurginus OR Panurgus OR Paradioxys OR Parammobatodes OR Pasites OR PANURGINAE OR Protosmia OR Pseudoanthidium OR Rhodanthidium OR RHOPHITINAE OR Rhophitoides OR Rophites OR Schmiedeknechtia OR Simpanurgus OR Sphecodes OR Stelis OR Stenoheriades OR Systropha OR Tarsalia OR Tetralonia OR Tetraloniella OR Thrincohalictus OR Thyreus OR Trachusa OR Triepeolus OR Xylocopa OR XYLOCOPINAE )</p> <p>Indexes=BCI Timespan=All years</p>	

### CAB Abstracts

Set	Query	Comments
# 14	#13 AND #4 Indexes=CAB Abstracts Timespan=All years	Q4
# 13	#12 OR #11 Indexes=CAB Abstracts Timespan=All years	
# 12	TI=( ((behavior* OR behaviour* OR distance* OR time* OR length* OR duration* OR performance* OR abilit*) AND (fly* OR flight* OR trip OR trips)) OR ((forag* OR collect* OR search*) AND ( fly* OR flight* OR trip OR trips) ) OR (activit* AND (forag* OR monitor* OR track*))) Indexes=CAB Abstracts Timespan=All years	
# 11	TS=(( (behavior* OR behaviour* OR distance* OR time* OR length* OR duration* OR performance* OR abilit*) NEAR/5 (fly* OR flight* OR trip OR trips)) OR ((forag* OR collect* OR search*) NEAR/5 ( fly* OR flight* OR trip OR trips) ) OR (activit* NEAR/5 (forag* OR monitor* OR track*))) Indexes=CAB Abstracts Timespan=All years	
# 10	#9 AND #4 Indexes=CAB Abstracts Timespan=All years	Q3
# 9	TS=(energ* AND (collection* OR consumption OR cost OR costs OR demand* OR efficienc* OR forag* OR intak* OR need* OR performance )) Indexes=CAB Abstracts Timespan=All years	
# 8	#7 AND #4 Indexes=CAB Abstracts Timespan=All years	Q1 - Q2
# 7	#6 OR #5 Indexes=CAB Abstracts Timespan=All years	
# 6	TS=(( nectar* OR pollen* OR sugar OR carbohydrate*) NEAR/5 ("consume" OR "consumed" OR "consumes" OR consumption* OR intak* OR feed*) OR (food NEAR/5	

	("consume" OR "consumed" OR "consumes" OR consumption*) OR "food intake*") Indexes=CAB Abstracts Timespan=All years	
# 5	TI=((food* OR nectar* OR pollen* OR sugar OR carbohydrate*) AND ("consume" OR "consumed" OR "consumes" OR consumption* OR intak* OR feed* OR diet OR diets OR dietar*)) Indexes=CAB Abstracts Timespan=All years	
# 4	#3 OR #2 OR #1 Indexes=CAB Abstracts Timespan=All years	
# 3	TS=((Europe* OR Euroregion* OR "Euro region*" OR Austria* OR Belgium OR Belgian OR Bulgaria OR Bulgarian OR Croat* OR Cyprus OR Cypriot* OR Czech* OR Denmark* OR Danish OR Estonia* OR Finland OR finnish* OR France OR French* OR German* OR Greece OR "hellenic republic" OR Greek* OR Hungar* OR Ireland OR Irish OR Ital* OR Latvia OR Lithuania* OR Luxembourg* OR Malta OR maltese* OR Netherlands* OR dutch* OR Poland* OR Polish* OR Portug* OR Romania* OR Slovak* OR Slovenia* OR Spain OR Spanish* OR Swed* OR "United Kingdom" OR Britain OR British OR England* OR English* OR Scotland* OR Scottish OR Wales OR Welsh OR Liechtenstein* OR Iceland* OR Norway* OR Norwegian* OR Switzerland* OR Swiss* OR Albanian* OR Macedonia* OR Montenegro* OR Serbia* OR Bosnia* OR Kosov* OR Benelux OR Czechoslovakia* OR Scandinav* OR Yugoslavia* OR Balkan* OR "Baltic countr*" OR "Baltic state*" OR "Iberian peninsula" OR Iberia OR "Mediterranean countr*" OR "Mediterranean state*" OR "Mediterranean region*" OR "Nordic countr*" OR "Nordic state*") AND (bee OR bees)) Indexes=CAB Abstracts Timespan=All years	
# 2	TS=("managed bee" OR "managed bees" OR "Apis mellifera" OR "Apis mellifica" OR "a mellifera" OR "a mellifica" OR Honeybee* OR "honey bee*" OR Bombus OR Bumblebee* OR "Bumble bee*" OR "B. alpinus " OR "B. argillaceus " OR "B. armeniacus " OR "B. balteatus " OR "B. barbutellus " OR "B. bohemicus " OR "B. brodmannicus " OR "B. campestris" OR "B. cingulatus " OR "B. confusus " OR "B. consobrinus" OR "B. cryptarum" OR "B. cullumanus " OR "B. deuteronymus" OR "B. distinguendus" OR "B. flavidus " OR "B. fragrans " OR "B. gerstaeckeri " OR "B. glacialis " OR "B. haematurus " OR "B. hortorum " OR "B. humilis " OR "B. hyperboreus " OR "B. hypnorum " OR "B. inexpectatus " OR "B. jonellus" OR "B. laesus" OR "B. lapidarius" OR "B. lapponicus" OR "B. lucorum" OR "B. magnus" OR "B. mendax" OR "B. mesomelas" OR "B. mlokosievitzii" OR "B. mocsaryi" OR "B. modestus" OR "B. monticola" OR "B. mucidus" OR "B. muscorum" OR "B. niveatus" OR "B. norvegicus" OR "B. pascuorum" OR "B. patagiatus" OR "B. perezi" OR "B. pereziellus" OR "B. polaris" OR "B. pomorum" OR "B. pratorum" OR "B. pyrenaeus" OR "B. quadricolor" OR "B. reinigiellus" OR "B. ruderarius" OR "B. ruderatus" OR "B. rupestris" OR "B. saltuarius" OR "B. schrencki" OR "B. semenoviellus" OR "B. sichelii" OR "B. soroensis" OR "B. sporadicus" OR "B. subterraneus" OR "B. sylvaram" OR "B. sylvestris" OR "B. terrestris" OR "B. vestalis" OR "B. veteranus" OR "B. wurflenii" OR "B. zonatus") Indexes=CAB Abstracts Timespan=All years	
# 1	TS=((solitary NEAR/3 (bee OR bees)) OR Afranthidium OR Aglaopis OR Amegilla OR Ammobates OR Ammobatoides OR Ancyla OR Andrena OR ANDRENIDAE OR ANDRENINAE OR Anthidiellum OR Anthidium OR Anthophora OR APIDAE OR APINAE or Biastes OR Camptopoeum OR Ceratina OR Ceylalictus OR Chelostoma OR Chiasmognathus OR Clavipanurgus OR Coelioxys OR Colletes OR COLLETIDAE OR COLLETINAE OR Cubitalia OR Dasypoda OR DASYPODAINAE OR Dioxys OR Dufourea OR Ensliniana OR Eoanthidium OR Epeoloides OR Epeolus OR Eucera OR Flavipanurgus OR Habropoda OR Haetosmia OR HALICTIDAE OR HALICTINAE OR Halictus OR Heriades OR Hofferia OR Hoplitis OR HYLAENIAE OR Hylaeus OR Icteranthidium OR Lasioglossum OR Lithurgus OR Macropis OR Megachile OR MEGACHILIDAE OR MEGACHILINAE OR Melecta OR Melitta OR MELITTIDAE OR MELITTINAE OR Melitturga OR Metadioxys OR (Nomada AND (bee OR bees)) OR NOMADINAE OR NOMIINAE OR Nomiapis OR NOMIOIDINAE OR Nomioides OR Osmia OR Panurginus OR Panurgus OR Paradioxys OR Parammobatodes OR Pasites OR PANURGINAE OR Protosmia OR Pseudoanthidium OR Rhodanthidium OR RHOPHITINAE OR Rophitoides OR Rophites OR Schmiedeknechtia OR Simpanurgus OR Sphecodes OR Stelis OR Stenoheriades OR Systropha OR Tarsalia OR Tetralonia OR Tetraloniella OR Thrincohalictus OR Thyreus OR Trachusa OR Triepeolus OR Xylocopa OR XYLOCOPINAE ) Indexes=CAB Abstracts Timespan=All years	

## Scopus

Set	Query	Comments
14	#13 AND #4	Q4
13	#11 OR #12	
12	TITLE((( OR OR OR OR OR OR OR ) AND ( OR OR OR )) OR (( OR OR ) AND ( OR OR OR )) OR ( AND ( OR OR )))	
11	TITLE-ABS-KEY((( OR OR OR OR OR OR OR ) W/5 ( OR OR OR )) OR ( ( OR OR ) W/5 ( OR OR OR )) OR ( W/5 ( OR OR )))	
10	#4 AND #9	Q3
9	TITLE-ABS-KEY( AND ( OR OR OR OR OR OR OR OR OR ))	
8	#4 AND #7	Q1 – Q2
7	#6 OR #5	
6	TITLE(( food* OR nectar* OR pollen* OR sugar OR carbohydrate*) AND ( consume OR consumed OR consumes OR consumption* OR intak* OR feed* OR diet OR diets OR dietar*))	
5	TITLE-ABS-KEY(( food* OR nectar* OR pollen* OR sugar OR carbohydrate*) W/5 ( consume OR consumed OR consumes OR consumption* OR intak* OR feed* OR diet OR diets OR dietar*))	
4	#1 OR #2 OR #3	
3	TITLE-ABS-KEY(( solitary W/3 ( bee OR bees )) OR afranthidium OR aglaoapis OR amegilla OR ammobates OR ammobatoides OR ancyla OR andrena OR andrenidae OR andreninae OR anthidiellum OR anthidium OR anthophora OR apidae OR apinae OR biastes OR camptopoeum OR ceratina OR ceylalictus OR chelostoma OR chiasmognathus OR clavipanurgus OR coelioxys OR colletes OR colletidae OR colletinae OR cubitalia OR dasyпода OR dasyподainae OR {dioxys} OR dufourea OR encliniana OR eoanthidium OR epeoloides OR epeolus OR eucera OR flavipanurgus OR habropoda OR haetosmia OR halictidae OR halictinae OR halictus OR heriades OR hofferia OR hoplitis OR hylaenae OR hylaeus OR icteranthidium OR lasioglossum OR lithurgus OR macropis OR megachile OR megachilidae OR megachilinae OR melecta OR melitta OR melittidae OR melittinae OR melitturga OR metadioxys OR ( nomada AND ( bee OR bees )) OR nomadinae OR nomiinae OR nomiapis OR nomioidinae OR nomioides OR osmia OR panurginus OR panurgus OR paradiioxys OR parammobatodes OR pasites OR panurginae OR protosmia OR pseudoanthidium OR rhodanthidium OR rhophitinae OR rhophitoides OR rophites OR schmiedeknechtia OR simpanurgus OR sphecodes OR stelis OR stenoheria OR systropha OR tarsalia OR tetralonia OR tetraloniella OR thrincohalictus OR thyreus OR trachusa OR triepeolus OR xylocopa OR xylocopinae )	
2	TITLE-ABS-KEY( "managed bee" OR "managed bees" OR "Apis mellifera" OR "Apis mellifica" OR "a mellifera" OR "a mellifica" OR honeybee* OR "honey bee*" OR bombus OR bumblebee* OR "Bumble bee*" OR "B. alpinus " OR "B. argillaceus " OR "B. armeniacus " OR "B. balteatus " OR "B. barbutellus " OR "B. bohemicus " OR "B. brodmannicus " OR "B. campestris" OR "B. cingulatus " OR "B. confusus " OR "B. consobrinus" OR "B. cryptarum" OR "B. cullumanus " OR "B. deuteronymus" OR "B. distinguendus" OR "B. flavidus " OR "B. fragrans " OR "B. gerstaeckeri " OR "B. glacialis " OR "B. haematurus " OR "B. hortorum " OR "B. humilis " OR "B. hyperboreus " OR "B. hypnorum " OR "B. inexpectatus " OR "B. jonellus" OR "B. laesus" OR "B. lapidarius" OR "B. lapponicus" OR "B. lucorum" OR "B. magnus" OR "B. mendax" OR "B. mesomelas" OR "B. mlokosievitzii" OR "B. mocsaryi" OR "B. modestus" OR "B. monticola" OR "B. mucidus" OR "B. muscorum" OR "B. niveatus" OR "B. norvegicus" OR "B. pascuorum" OR "B. patagiatus" OR "B. perezi" OR "B. pereziellus" OR "B. polaris" OR "B. pomorum" OR "B. pratorum" OR "B. pyrenaicus" OR "B. quadricolor" OR "B. reinigiellus" OR "B. ruderarius" OR "B. ruderatus" OR "B. rupestris" OR "B. saltuarius" OR "B. schrencki" OR "B. semenoviellus" OR "B. sichelii" OR "B. soroensis" OR "B. sporadicus" OR "B. subterraneus" OR "B. sylvarum" OR "B. sylvestris" OR "B. terrestris" OR "B. vestalis" OR "B. veteranus" OR "B. wurflenii" OR "B. zonatus" )	
1	( TITLE-ABS-KEY( ( europe* OR euroregion* OR "Euro region*" OR austria* OR belgium OR belgian OR bulgaria OR bulgarian OR croatia* OR cyprus OR cyriot* OR czech* OR denmark* OR danish OR estonia* OR	

finland OR finnish\* OR france OR french\* OR german\* OR greece OR "hellenic republic" OR greek\* OR hungar\* OR ireland OR irish OR ital\* OR latvia OR lithuania\* OR luxembourg\* OR malta OR maltese\* OR netherlands\* OR dutch\* OR poland\* OR polish\* OR portug\* OR romania\* OR slovak\* OR slovenia\* OR spain OR spanish\* OR swed\* OR "United Kingdom" OR britain OR british OR england\* OR english\* OR scotland\* OR scottish OR wales OR welsh OR liechtenstein\* OR iceland\* OR norway\* OR norwegian\* OR switzerland\* OR swiss\* OR albanian\* OR macedonia\* OR montenegro\* OR serbia\* OR bosnia\* OR kosov\* OR benelux OR czechoslovakia\* OR scandinav\* OR yugoslavia\* OR balkan\* OR "Baltic countr\*" OR "Baltic state\*" OR "Iberian peninsula" OR iberia OR "Mediterranean countr\*" OR "Mediterranean state\*" OR "Mediterranean region\*" OR "Nordic countr\*" OR "Nordic state\*" ) AND ( bee OR bees ) )

## Web of Science

Set	Query	Comments
# 14	#13 AND #4 Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	Q4
# 13	#12 OR #11 Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	
# 12	TI=((behavior* OR behaviour* OR distance* OR time* OR length* OR duration* OR performance* OR abilit*) AND (fly* OR flight* OR trip OR trips)) OR ((forag* OR collect* OR search*) AND ( fly* OR flight* OR trip OR trips ) OR (activit* AND (forag* OR monitor* OR track*))) Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	
# 11	TS=((behavior* OR behaviour* OR distance* OR time* OR length* OR duration* OR performance* OR abilit*) NEAR/5 (fly* OR flight* OR trip OR trips)) OR ((forag* OR collect* OR search*) NEAR/5 ( fly* OR flight* OR trip OR trips ) OR (activit* NEAR/5 (forag* OR monitor* OR track*))) Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	
# 10	#9 AND #4 Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	Q3
# 9	TS=(energ* AND (collection* OR consumption OR cost OR costs OR demand* OR efficienc* OR forag* OR intak* OR need* OR performance )) Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	
# 8	#7 AND #4 Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	Q1 - Q2
# 7	#6 OR #5 Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	
# 6	TI=((food* OR nectar* OR pollen* OR sugar OR carbohydrate*) AND ("consume" OR "consumed" OR "consumes" OR consumption* OR intak* OR feed* OR diet OR diets OR dietar*)) Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	
# 5	TS=((food* OR nectar* OR pollen* OR sugar OR carbohydrate*) NEAR/5 ("consume" OR "consumed" OR "consumes" OR consumption* OR intak* OR feed* OR diet OR diets OR dietar*)) Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	
# 4	#3 OR #2 OR #1 Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	
# 3	TS=((Europe* OR Euroregion* OR "Euro region*" OR Austria* OR Belgium OR Belgian OR Bulgaria OR Bulgarian OR Croat* OR Cyprus OR Cypriot* OR Czech* OR Denmark* OR Danish OR Estonia* OR Finland OR finnish* OR France OR French* OR German* OR Greece OR "hellenic republic" OR Greek* OR Hungar* OR Ireland OR Irish OR Ital* OR Latvia OR Lithuania* OR Luxembourg* OR Malta OR maltese* OR Netherlands* OR dutch* OR Poland* OR Polish* OR Portug* OR Romania* OR Slovak* OR Slovenia* OR Spain OR Spanish* OR Swed* OR "United Kingdom" OR Britain OR British OR England* OR English* OR Scotland* OR Scottish OR Wales OR Welsh OR Liechtenstein* OR Iceland* OR Norway* OR Norwegian* OR Switzerland* OR Swiss* OR Albanian* OR Macedonia* OR Montenegro* OR Serbia* OR Bosnia* OR Kosov* OR Benelux OR Czechoslovakia* OR Scandinav* OR Yugoslavia* OR Balkan* OR "Baltic countr*" OR "Baltic state*" OR "Iberian peninsula" OR Iberia OR "Mediterranean countr*" OR	

	"Mediterranean state*" OR "Mediterranean region*" OR "Nordic countr*" OR "Nordic state*") AND (bee OR bees)) Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	
# 2	TS=("managed bee" OR "managed bees" OR "Apis mellifera" OR "Apis mellifica" OR "a mellifera" OR "a mellifica" OR Honeybee* OR "honey bee*" OR Bombus OR Bumblebee* OR "Bumble bee*" OR "B. alpinus " OR "B. argillaceus " OR "B. armeniacus " OR "B. balteatus " OR "B. barbutellus " OR "B. bohemicus " OR "B. brodmannicus " OR "B. campestris" OR "B. cingulatus " OR "B. confusus " OR "B. consobrinus" OR "B. cryptarum" OR "B. cullumanus " OR "B. deuteronymus" OR "B. distinguendus" OR "B. flavidus " OR "B. fragrans " OR "B. gerstaeckeri " OR "B. glacialis " OR "B. haematurus " OR "B. hortorum " OR "B. humilis " OR "B. hyperboreus " OR "B. hypnorum " OR "B. inexpectatus " OR "B. jonellus" OR "B. laesus" OR "B. lapidarius" OR "B. lapponicus" OR "B. lucorum" OR "B. magnus" OR "B. mendax" OR "B. mesomelas" OR "B. mlokosievitzii" OR "B. mocsaryi" OR "B. modestus" OR "B. monticola" OR "B. mucidus" OR "B. muscorum" OR "B. niveatus" OR "B. norvegicus" OR "B. pascuorum" OR "B. patagiatus" OR "B. perezi" OR "B. pereziellus" OR "B. polaris" OR "B. pomorum" OR "B. pratorum" OR "B. pyrenaicus" OR "B. quadricolor" OR "B. reinigiellus" OR "B. ruderarius" OR "B. ruderatus" OR "B. rupestris" OR "B. saltuarius" OR "B. schrencki" OR "B. semenoviellus" OR "B. sichelii" OR "B. soroensis" OR "B. sporadicus" OR "B. subterraneus" OR "B. sylvarum" OR "B. sylvestris" OR "B. terrestris" OR "B. vestalis" OR "B. veteranus" OR "B. wurflenii" OR "B. zonatus") Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	
# 1	TS=((solitary NEAR/3 (bee OR bees)) OR Afranthidium OR Aglaopis OR Amegilla OR Ammobates OR Ammobatoides OR Ancyla OR Andrena OR ANDRENIDAE OR ANDRENINAE OR Anthidiellum OR Anthidium OR Anthophora OR APIDAE OR APINAE or Biastes OR Camptopoeum OR Ceratina OR Ceylalictus OR Chelostoma OR Chiasmognathus OR Clavipanurgus OR Coelioxys OR Colletes OR COLLETIDAE OR COLLETINAE OR Cubitalia OR Dasygona OR DASYPODAINAE OR Dioxys OR Dufourea OR Ensliniana OR Eoanthidium OR Epeoloides OR Epeolus OR Eucera OR Flavipanurgus OR Habropoda OR Haetosmia OR HALICTIDAE OR HALICTINAE OR Halictus OR Heriades OR Hofferia OR Hoplitis OR HYLAENIAE OR Hylaeus OR Icteranthidium OR Lasioglossum OR Lithurgus OR Macropis OR Megachile OR MEGACHILIDAE OR MEGACHILINAE OR Melecta OR Melitta OR MELITTIDAE OR MELITTINAE OR Melitturga OR Metadioxys OR (Nomada AND (bee OR bees)) OR NOMADINAE OR NOMIINAE OR Nomiapis OR NOMIOIDINAE OR Nomioides OR Osmia OR Panurginus OR Panurgus OR Paradioxys OR Parammobatodes OR Pasites OR PANURGINAE OR Protosmia OR Pseudoanthidium OR Rhodanthidium OR RHOPHITINAE OR Rhophitoides OR Rophites OR Schmiedeknechtia OR Simpanurgus OR Sphecodes OR Stelis OR Stenoheriades OR Systropha OR Tarsalia OR Tetralonia OR Tetraloniella OR Thrincohalictus OR Thyreus OR Trachusa OR Triepeolus OR Xylocopa OR XYLOCOPINAE ) Indexes=SCI-EXPANDED, CPCI-S, ESCI Timespan=All years	

## Zoological Record

Set	Query	Comments
# 14	#13 AND #4 Indexes=Zoological Record Timespan=All years	Q4 foraging behaviour
# 13	#12 OR #11 Indexes=Zoological Record Timespan=All years	
# 12	TI=((behavior* OR behaviour* OR distance* OR time* OR length* OR duration* OR performance* OR abilit*) AND (fly* OR flight* OR trip OR trips)) OR ((forag* OR collect* OR search*) AND ( fly* OR flight* OR trip OR trips ) OR (activit* AND (forag* OR monitor* OR track*))) Indexes=Zoological Record Timespan=All years	
# 11	TS=((behavior* OR behaviour* OR distance* OR time* OR length* OR duration* OR performance* OR abilit*) NEAR/5 (fly* OR flight* OR trip OR trips)) OR ((forag* OR collect* OR search*) NEAR/5 ( fly* OR flight* OR trip OR trips ) OR (activit* NEAR/5 (forag* OR monitor* OR track*))) Indexes=Zoological Record Timespan=All years	
# 10	#9 AND #4 Indexes=Zoological Record Timespan=All years	Q3 energetics
# 9	TS=(energ* AND (collection* OR consumption OR cost OR costs OR demand* OR efficienc* OR forag* OR intak* OR need* OR performance ))	

# 8	Indexes=Zoological Record Timespan=All years #7 AND #4 Indexes=Zoological Record Timespan=All years	Q1 and 2 food consumption
# 7	#6 OR #5 Indexes=Zoological Record Timespan=All years	
# 6	TI=((food* OR nectar* OR pollen* OR sugar OR carbohydrate*) AND ("consume" OR "consumed" OR "consumes" OR consumption* OR intak* OR feed* OR diet OR diets OR dietar*)) Indexes=Zoological Record Timespan=All years	
# 5	TS=((food* OR nectar* OR pollen* OR sugar OR carbohydrate*) NEAR/5 ("consume" OR "consumed" OR "consumes" OR consumption* OR intak* OR feed* OR diet OR diets OR dietar*)) Indexes=Zoological Record Timespan=All years	
# 4	#3 OR #2 OR #1 Indexes=Zoological Record Timespan=All years	
# 3	TS=((Europe* OR Euroregion* OR "Euro region*" OR Austria* OR Belgium OR Belgian OR Bulgaria OR Bulgarian OR Croat* OR Cyprus OR Cypriot* OR Czech* OR Denmark* OR Danish OR Estonia* OR Finland OR finnish* OR France OR French* OR German* OR Greece OR "hellenic republic" OR Greek* OR Hungar* OR Ireland OR Irish OR Ital* OR Latvia OR Lithuania* OR Luxembourg* OR Malta OR maltese* OR Netherlands* OR dutch* OR Poland* OR Polish* OR Portug* OR Romania* OR Slovak* OR Slovenia* OR Spain OR Spanish* OR Swed* OR "United Kingdom" OR Britain OR British OR England* OR English* OR Scotland* OR Scottish OR Wales OR Welsh OR Liechtenstein* OR Iceland* OR Norway* OR Norwegian* OR Switzerland* OR Swiss* OR Albanian* OR Macedonia* OR Montenegro* OR Serbia* OR Bosnia* OR Kosov* OR Benelux OR Czechoslovakia* OR Scandinav* OR Yugoslavia* OR Balkan* OR "Baltic countr*" OR "Baltic state*" OR "Iberian peninsula" OR Iberia OR "Mediterranean countr*" OR "Mediterranean state*" OR "Mediterranean region*" OR "Nordic countr*" OR "Nordic state*" ) AND (bee OR bees)) Indexes=Zoological Record Timespan=All years	
# 2	TS=("managed bee" OR "managed bees" OR "Apis mellifera" OR "Apis mellifica" OR "a mellifera" OR "a mellifica" OR Honeybee* OR "honey bee*" OR Bombus OR Bumblebee* OR "Bumble bee*" OR "B. alpinus " OR "B. argillaceus " OR "B. armeniacus " OR "B. balteatus " OR "B. barbutellus " OR "B. bohemicus " OR "B. brodmannicus " OR "B. campestris" OR "B. cingulatus " OR "B. confusus " OR "B. consobrinus" OR "B. cryptarum" OR "B. cullumanus " OR "B. deuteronymus" OR "B. distinguendus" OR "B. flavidus " OR "B. fragrans " OR "B. gerstaeckeri " OR "B. glacialis " OR "B. haematurus " OR "B. hortorum " OR "B. humilis " OR "B. hyperboreus " OR "B. hypnorum " OR "B. inexpectatus " OR "B. jonellus" OR "B. laesus" OR "B. lapidarius" OR "B. lapponicus" OR "B. lucorum" OR "B. magnus" OR "B. mendax" OR "B. mesomelas" OR "B. mlokosievitzii" OR "B. mocsaryi" OR "B. modestus" OR "B. monticola" OR "B. mucidus" OR "B. muscorum" OR "B. niveatus" OR "B. norvegicus" OR "B. pascuorum" OR "B. patagiatus" OR "B. perezi" OR "B. pereziellus" OR "B. polaris" OR "B. pomorum" OR "B. pratorum" OR "B. pyrenaeus" OR "B. quadricolor" OR "B. reinigiellus" OR "B. ruderarius" OR "B. ruderatus" OR "B. rupestris" OR "B. saltuarius" OR "B. schrencki" OR "B. semenoviellus" OR "B. sichelii" OR "B. soroeensis" OR "B. sporadicus" OR "B. subterraneus" OR "B. sylvorum" OR "B. sylvestris" OR "B. terrestris" OR "B. vestalis" OR "B. veteranus" OR "B. wurflenii" OR "B. zonatus") Indexes=Zoological Record Timespan=All years	
# 1	TS=((solitary NEAR/3 (bee OR bees)) OR Afranthidium OR Aglaoapis OR Amegilla OR Ammobates OR Ammobatoides OR Ancyla OR Andrena OR ANDRENIDAE OR ANDRENINAE OR Anthidiellum OR Anthidium OR Anthophora OR APIDAE OR APINAE or Biastes OR Camptopoeum OR Ceratina OR Ceylalictus OR Chelostoma OR Chiasmognathus OR Clavipanurgus OR Coelioxys OR Colletes OR COLLETIDAE OR COLLETINAE OR Cubitalia OR Dasypoda OR DASYPODAINAE OR Dioxys OR Dufourea OR Ensliniana OR Eoanthidium OR Epeoloides OR Epeolus OR Eucera OR Flavipanurgus OR Habropoda OR Haetosmia OR HALICTIDAE OR HALICTINAE OR Halictus OR Heriades OR Hofferia OR Hoplitis OR HYLAEINAE OR Hylaeus OR Icteranthidium OR Lasioglossum OR Lithurgus OR Macropis OR Megachile OR MEGACHILIDAE OR MEGACHILINAE OR Melecta OR Melitta OR MELITTIDAE OR MELITTINAE OR Melitturga OR Metadioxys OR (Nomada AND (bee OR bees)) OR NOMADINAE OR NOMIINAE OR Nomiapis OR NOMIOIDINAE OR Nomioides OR Osmia OR Panurginus OR Panurgus OR Paradioxys OR Parammobatodes OR Pasites OR PANURGINAE OR Protosmia OR Pseudoanthidium OR	



Rhodanthidium OR RHOPHITINAE OR Rhophitoides OR Rophites OR Schmiedeknechtia  
OR Simpanurgus OR Sphecodes OR Stelis OR Stenoheriades OR Systropha OR Tarsalia  
OR Tetralonia OR Tetraloniella OR Thrincohalictus OR Thyreus OR Trachusa OR  
Triepeolus OR Xylocopa OR XYLOCOPINAE )  
Indexes=Zoological Record Timespan=All years

## Appendix D – Search strings on sugar content

### Biosis

Set	Query
# 6	#5 AND #4 AND #3 Indexes=BCI Timespan=All years
# 5	#2 OR #1 Indexes=BCI Timespan=All years
# 4	TS=( (sugar OR sugars) NEAR/7 (composition* OR concentration* OR content OR contents OR volume*)) Indexes=BCI Timespan=All years
# 3	TS=(nectar OR nectars) Indexes=BCI Timespan=All years
# 2	TS=(Alfalfa OR Medicago OR "M sativa" OR almond OR almonds OR Prunus OR "P amygdalus" OR "P communis" OR Amygdalus OR "A communis" OR anise OR Pimpinella OR "P anisum" OR badian* OR Illicium OR "I verum" OR carawa* OR Carum OR "C carvi" OR coriander* OR Coriandrum OR "C sativum" OR OR cumin* OR "C cyminum" OR fennel OR Foeniculum OR Juniperus OR "juniper berr*" OR "J communis" OR apple* OR Malus OR "M pumila" OR "M Sylvestris" OR "P sylvestris" OR "M communis" OR apricot* OR Pyrus OR "P malus" OR "P armeniaca" OR Artichoke* OR Cynara OR "C scolymus" OR asparagus OR "A officianils" OR Avocado* OR Persea OR "P Americana" OR Banana* OR Musa OR "M sapientum" OR "M cavendishii" OR "M nana" OR Barley OR Hordeum OR "H disticum" OR "H vulgare" OR Phaseolus OR blueberr* OR "wild bilberr*" OR whortleberr* OR Vaccinium OR "V myrtillus" OR "V Corymbosum" OR cranberr* OR "myrtle berr*" OR bean OR beans OR faba OR Buckwheat* OR Fagopyrum OR Fagopyron OR "F esculentum" OR Cabbage* OR Brassica OR "B oleracea" OR "pak choi" OR pakchoi OR "B chinensis" OR "brussels sprout*" OR collard* OR kale OR kales OR kohlrabi* OR Carob OR carobs OR Ceratonia OR "C siliqua" OR Carrot* OR Daucus OR "D carota" OR "Castor oil seed*" OR "castor oil plant*" OR Ricinus OR "R communis" OR Cauliflower* OR broccoli* OR Cherry OR cherries OR "P avium" OR Cerasus OR "C avium" OR Chestnut* OR Castanea OR "C vesca" OR "C vulgaris" OR "C sativa" OR "Chick pea*" OR chickpea* OR Cicer OR "C arietinum" OR Chicor* OR "C intybus" OR Chilli OR Chillies OR pepper* OR paprika* OR Capsicum OR "C frutescens" OR "C annuum" OR allspice* OR Pimenta OR "P officinalis" OR Clover* OR Trifolium OR Coffee* OR Coffea OR "C arabica" OR "C robusta" OR "C liberica" OR "Cow pea*" OR cowpea* OR "blackeye pea*" OR Vigna OR "V unguiculata" OR Cranberr* OR Vaccinium OR "V macrocarpon" OR "Voxycoccus" OR Cucumber* OR gherkin* OR Cucumis OR "c sativus" OR Currant* OR goosecurrant* OR Ribes OR "R nigrum" OR "R rubrum" OR Elder* OR Sambucus OR "S nigra" OR Garlic* OR Allium OR "A sativum" OR Gooseberr* OR "R grossularia" OR Grapefruit* OR pomelo* OR Citrus OR "C maxima" OR "C grandis" OR "C paradisi" OR Grapes OR Vitis OR "V vinifera" OR Bent OR bents OR redtop OR redtops OR "fiorin grass" OR Agrostis OR bluegrass OR poa OR "Columbus grass" OR Sorghum OR "S alnum" OR fescue OR Festuca OR napier OR "elephant grass" OR pennisetum OR "P purpureum" OR "orchad grass" OR Dactylis OR "D glomerata" OR "Rhodes grass" OR Chloris OR "C gayana" OR Hemp OR Cannabis OR "C sativa" OR Leek OR leeks OR "A porrum" OR chive OR chives OR "A schoenoprasum" OR Bridesfoot* OR trefoil* OR lotus OR "L corniculatus" OR lespedeza OR kudzu OR Prueraria OR "P lobata" OR sesbania OR sainfoin OR esparcette OR Onobrychis OR "O sativa" OR sulla OR Hedysarum OR "H coronarium" OR Lemon* OR lime* OR "C limon" OR "C aurantifolia" OR "C limetta" OR Lentil* OR Lens OR "L esculenta" OR "E lens" OR "L culinaris" OR Linseed* OR Linum OR "L usitatissimum" OR flaxseed* OR Lupins OR lupinus OR Melon* OR Cucumis OR "C melo" OR "Mustard seed*" OR "White mustard" OR "B alba" OR "B hirta" OR sinapis OR "black mustard" OR "B nigra" OR Onion* OR Okra OR okras OR Abelmoschus OR "A esculentus" OR Hibiscus OR "H esculentus" OR gombo OR Onion* OR "A cepa" OR Orange* OR "C sinensis" OR "C aurantium" OR Peach* OR nectarin* OR "P persica" OR "A persica" OR persica OR "P laevis" OR Pear* OR Pyrus OR "P communis" OR Pea OR peas OR Pisum OR "P sativum" OR "P arvense" OR Peppermint OR Mentha OR "M piperita" OR Persimmon* OR Diospyros OR "D kaki" OR "D virginiana" OR Plum OR plums OR greengage* OR Mirabelle* OR damson* OR "P domestica" OR Sloe* OR "P spinosa" OR Pumpkin* OR squash* OR gourd* OR Cucurbita OR marrow* OR Pyrethrum* OR Chrysanthemum OR "C cinerariifolium" OR Quince* OR Cydonia OR "C oblonga" OR "C vulgaris" OR "C japonica" OR Rapeseed* OR "B napus" OR Raspberr* OR Rubus OR "R idaeus" OR blackberr* OR mulberr* OR loganberr* OR Safflower* OR Carthamus OR "C tinctorius" OR Cotton OR Gossypium OR Serradella* OR birdsfoot* OR Ornithopus OR "O sativus" OR Sesam* OR "S indicum" OR Soybean* OR Glycine OR "G max" OR OR soja OR soya OR "Bay leave*" OR Laurus OR "L nobilis" OR dill OR Anethum OR "A graveolens" OR fenugreek OR Trigonella OR saffron OR Crocus OR "C sativus" OR thyme OR thymus OR turmeric OR Curcuma OR Spinach* OR Spinacia OR "S oleracea" OR Tetragonia OR "T espansa" OR Artiplex OR "A hortensis" OR Strawberr* OR Fragaria OR "Sugar beet*" OR Beta OR "B vulgaris" OR Sunflower* OR Helianthus OR "H annuus" OR "Sweet potato*" OR Ipomoea OR batata* OR "I batatas" OR Tangerine* OR mandarin* OR clementine* OR "c reticulata" OR satsuma OR "C unshiu" OR Turnip* OR "B rapa" OR Vetch* OR Vicia OR "V sativa" OR "Vipers grass" OR Scorzonera OR "S hispanica" OR Walnut* OR Juglans OR "J regia" OR Watermelon* OR Citrullus OR "C

	vulgaris")Indexes=BCI Timespan=All years
# 1	TS=(( crop OR crops OR flower* OR floral OR fruit* OR *bee OR *bees OR bee OR bees)) Indexes=BCI Timespan=All years

## CAB Abstracts

Set	Query
# 6	#5 AND #4 AND #3 Indexes=CAB Abstracts Timespan=All years
# 5	TS=( sugar OR sugars) NEAR/7 (composition* OR concentration* OR content OR contents OR volume*) Indexes=CAB Abstracts Timespan=All years
# 4	TS=(nectar OR nectars) Indexes=CAB Abstracts Timespan=All years
# 3	#2 OR #1 Indexes=CAB Abstracts Timespan=All years
# 2	TS=(Alfalfa OR Medicago OR "M sativa" OR almond OR almonds OR Prunus OR "P amygdalus" OR "P communis" OR Amygdalus OR "A communis" OR anise OR Pimpinella OR "P anisum" OR badian* OR Illicium OR "I verum" OR carawa* OR Carum OR "C carvi" OR coriander* OR Coriandrum OR "C sativum" OR cumin* OR "C cyminum" OR fennel OR Foeniculum OR Juniperus OR "juniper berr*" OR "J communis" OR apple* OR Malus OR "M pumila" OR "M Sylvestris" OR "P sylvestris" OR "M communis" OR apricot* OR Pyrus OR "P malus" OR "P armeniaca" OR Artichoke* OR Cynara OR "C scolymus" OR asparagus OR "A officianils" OR Avocado* OR Persea OR "P Americana" OR Banana* OR Musa OR "M sapientum" OR "M cavendishii" OR "M nana" OR Barley OR Hordeum OR "H disticum" OR "H vulgare" OR Phaseolus OR blueberr* OR "wild bilberr*" OR whortleberr* OR Vaccinium OR "V myrtillus" OR "V Corymbosum" OR cranberr* OR "myrtle berr*" OR bean OR beans OR faba OR Buckwheat* OR Fagopyrum OR Fagopyron OR "F esculentum" OR Cabbage* OR Brassica OR "B oleracea" OR "pak choi" OR pakchoi OR "B chinensis" OR "brussels sprout*" OR collard* OR kale OR kales OR kohlrabi* OR Carob OR carobs OR Ceratonia OR "C siliqua" OR Carrot* OR Daucus OR "D carota" OR "Castor oil seed*" OR "castor oil plant*" OR Ricinus OR "R communis" OR Cauliflower* OR broccoli* OR Cherry OR cherries OR "P avium" OR Cerasus OR "C avium" OR Chestnut* OR Castanea OR "C vesca" OR "C vulgaris" OR "C sativa" OR "Chick pea*" OR chickpea* OR Cicer OR "C arietinum" OR Chicor* OR "C intybus" OR Chilli OR Chillies OR pepper* OR paprika* OR Capsicum OR "C frutescens" OR "C annuum" OR allspice* OR Pimenta OR "P officinalis" OR Clover* OR Trifolium OR Coffee* OR Coffea OR "C arabica" OR "C robusta" OR "C liberica" OR "Cow pea*" OR cowpea* OR "blackeye pea*" OR Vigna OR "V unguiculata" OR Cranberr* OR Vaccinium OR "V macrocarpon" OR "Voxycoccus" OR Cucumber* OR gherkin* OR Cucumis OR "c sativus" OR Currant* OR goosecurrant* OR Ribes OR "R nigrum" OR "R rubrum" OR Elder* OR Sambucus OR "S nigra" OR Garlic* OR Allium OR "A sativum" OR Gooseberr* OR "R grossularia" OR Grapefruit* OR pomelo* OR Citrus OR "C maxima" OR "C grandis" OR "C paradisi" OR Grapes OR Vitis OR "V vinifera" OR Bent OR bents OR redtop OR redtops OR "fiorin grass" OR Agrostis OR bluegrass OR poa OR "Columbus grass" OR Sorghum OR "S alnum" OR fescue OR Festuca OR napier OR "elephant grass" OR pennisetum OR "P purpureum" OR "orchad grass" OR Dactylis OR "D glomerata" OR "Rhodes grass" OR Chloris OR "C gayana" OR Hemp OR Cannabis OR "C sativa" OR Leek OR leeks OR "A porrum" OR chive OR chives OR "A schoenoprasum" OR Birdsfoot* OR trefoil* OR lotus OR "L corniculatus" OR lespedeza OR kudzu OR Pueraria OR "P lobata" OR sesbania OR sainfoin OR esparcette OR Onobrychis OR "O sativa" OR sulla OR Hedysarum OR "H coronarium" OR Lemon* OR lime* OR "C limon" OR "C aurantifolia" OR "C limetta" OR Lentil* OR Lens OR "L esculenta" OR "E lens" OR "L culinaris" OR Linseed* OR Linum OR "L usitatissimum" OR flaxseed* OR Lupins OR lupinus OR Melon* OR Cucumis OR "C melo" OR "Mustard seed*" OR "White mustard" OR "B alba" OR "B hirta" OR sinapis OR "black mustard" OR "B nigra" OR Okra OR okras OR Abelmoschus OR "A esculentus" OR Hibiscus OR "H esculentus" OR gombo OR Onion* OR "A cepa" OR Orange* OR "C sinensis" OR "C aurantium" OR Peach* OR nectarin* OR "P persica" OR "A persica" OR persica OR "P laevis" OR Pear* OR Pyrus OR "P communis" OR Pea OR peas OR Pisum OR "P sativum" OR "P arvense" OR Peppermint OR Mentha OR "M piperita" OR Persimmon* OR Diospyros OR "D kaki" OR "D virginiana" OR Plum OR plums OR greengage* OR Mirabelle* OR damson* OR "P domestica" OR Sloe* OR "P spinosa" OR Pumpkin* OR squash* OR gourd* OR Cucurbita OR marrow* OR Pyrethrum* OR Chrysanthemum OR "C cinerariifolium" OR Quince* OR Cydonia OR "C oblonga" OR "C vulgaris" OR "C japonica" OR Rapeseed* OR "B napus" OR Raspberr* OR Rubus OR "R idaeus" OR blackberr* OR mulberr* OR loganberr* OR Safflower* OR Carthamus OR "C tinctorius" OR Cotton OR Gossypium OR Serradella* OR birdsfoot* OR Ornithopus OR "O sativus" OR Sesam* OR "S indicum" OR Soybean* OR Glycine OR "G max" OR soja OR soya OR "Bay leave*" OR Laurus OR "L nobilis" OR dill OR Anethum OR "A graveolens" OR fenugreek OR Trigonella OR saffron OR Crocus OR "C sativus" OR thyme OR thymus OR turmeric OR Curcuma OR Spinach* OR Spinacia OR "S oleracea" OR Tetragonia OR "T espansa" OR Artiplex OR "A hortensis" OR Strawberr* OR Fragaria OR "Sugar beet*" OR Beta OR "B vulgaris" OR Sunflower* OR Helianthus OR "H annuus" OR "Sweet potato*" OR Ipomoea OR batata* OR

	"I batatas" OR Tangerine* OR mandarin* OR clementine* OR "c reticulata" OR satsuma OR "C unshiu" OR Turnip* OR "B rapa" OR Vetch* OR Vicia OR "V sativa" OR "Vipers grass" OR Scorzonera OR "S hispanica" OR Walnut* OR Juglans OR "J regia" OR Watermelon* OR Citrullus OR "C vulgaris")Indexes=BCI Timespan=All years
# 1	TS=(( crop OR crops OR flower* OR floral OR fruit* OR *bee OR *bees OR bee OR bees)) Indexes=CAB Abstracts Timespan=All years

## Scopus

Set	Query
7	#4 AND #5 AND #6
6	TITLE-ABS-KEY ( ( sugar OR sugars ) W/7 ( composition* OR concentration* OR content OR contents OR volume* ) )
5	TITLE-ABS-KEY ( nectar OR nectars )
4	#1 OR #2
2	TITLE-ABS-KEY ( alfalfa OR medicago OR "M sativa" OR almond OR almonds OR prunus OR "P amygdalus" OR "P communis" OR amygdalus OR "A communis" OR anise OR pimpinella OR "P anisum" OR badian* OR illicium OR "I verum" OR carawa* OR carum OR "C carvi" OR coriander* OR coriandrum OR "C sativum" OR cumin* OR "C cyminum" OR fennel OR foeniculum OR juniperus OR "juniper berr*" OR "J communis" OR apple* OR malus OR "M pumila" OR "M Sylvestris" OR "P sylvestris" OR "M communis" OR apricot* OR pyrus OR "P malus" OR "P armeniaca" OR artichoke* OR cynara OR "C scolymus" OR asparagus OR "A officianils" OR avocado* OR perseas OR "P Americana" OR banana* OR musa OR "M sapientum" OR "M cavendishii" OR "M nana" OR barley OR hordeum OR "H disticum" OR "H vulgare" OR phaseolus OR blueberr* OR "wild bilberr*" OR whortleberr* OR vaccinium OR "V myrtillus" OR "V Corymbosum" OR cranberr* OR "myrtle berr*" OR bean OR beans OR faba OR buckwheat* OR fagopyrum OR fagopyron OR "F esculentum" OR cabbage* OR brassica OR "B oleracea" OR "pak choi" OR pakchoi OR "B chinensis" OR "brussels sprout*" OR collard* OR kale OR kales OR kohlrabi* OR carob OR carobs OR ceratonia OR "C siliqua" OR carrot* OR daucus OR "D carota" OR "Castor oil seed*" OR "castor oil plant*" OR ricinus OR "R communis" OR cauliflower* OR broccoli* OR cherry OR cherries OR "P avium" OR cerasus OR "C avium" OR chestnut* OR castanea OR "C vesca" OR "C vulgaris" OR "C sativa" OR "Chick pea*" OR chickpea* OR cicer OR "C arietinum" OR chicor* OR "C intybus" OR chilli OR chillies OR pepper* OR paprika* OR capsicum OR "C frutescens" OR "C annuum" OR allspice* OR pimenta OR "P officinalis" OR clover* OR trifolium OR coffee* OR coffea OR "C arabica" OR "C robusta" OR "C liberica" OR "Cow pea*" OR cowpea* OR "blackeye pea*" OR vigna OR "V unguiculata" OR cranberr* OR vaccinium OR "V macrocarpon" OR "Voxyccoccus" OR cucumber* OR gherkin* OR cucumis OR "c sativus" OR currant* OR goosecurrant* OR ribes OR "R nigrum" OR "R rubrum" OR elder* OR sambucus OR "S nigra" OR garlic* OR allium OR "A sativum" OR gooseberr* OR "R grossularia" OR grapefruit* OR pomelo* OR citrus OR "C maxima" OR "C grandis" OR "C paradisi" OR grapes OR vitis OR "V vinifera" OR bent OR bents OR redtop OR redtops OR "fiorin grass" OR agrostis OR bluegrass OR poa OR "Columbus grass" OR sorghum OR "S alnum" OR fescue OR festuca OR napier OR "elephant grass" OR pennisetum OR "P purpureum" OR "orchard grass" OR dactylis OR "D glomerata" OR "Rhodes grass" OR chloris OR "C gayana" OR hemp OR cannabis OR "C sativa" OR leek OR leeks OR "A porrum" OR chive OR chives OR "A schoenoprasum" OR bridsfoot* OR trefoil* OR lotus OR "L corniculatus" OR lespedeza OR kudzu OR prueraria OR "P lobata" OR sesbania OR sainfoin OR esparcette OR onobrychis OR "O sativa" OR sulla OR hedysarum OR "H coronarium" OR lemon* OR lime* OR "C limon" OR "C aurantifolia" OR "C limetta" OR lentil* OR lens OR "L esculenta" OR "E lens" OR "L culinaris" OR linseed* OR linum OR "L usitatissimum" OR flaxseed* OR lupins OR lupinus OR melon* OR cucumis OR "C melo" OR "Mustard seed*" OR "White mustard" OR "B alba" OR "B hirta" OR sinapis OR "black mustard" OR "B nigra" OR okra OR okras OR abelmoschus OR "A esculentus" OR hibiscus OR "H esculentus" OR gombo OR onion* OR "A cepa" OR orange* OR "C sinensis" OR "C aurantium" OR peach* OR nectarin* OR "P persica" OR "A persica" OR persica OR "P laevis" OR pear* OR pyrus OR "P communis" OR pea OR peas OR pisum OR "P sativum" OR "P

	<p>arvense" OR peppermint OR mentha OR "M piperita" OR persimmon* OR diospyros OR "D kaki" OR "D virginiana" OR plum OR plums OR greengage* OR mirabelle* OR damson* OR "P domestica" OR sloe* OR "P spinosa" OR pumpkin* OR squash* OR gourd* OR cucurbita OR marrow* OR pyrethrum* OR chrysanthemum OR "C cinerariifolium" OR quince* OR cydonia OR "C oblonga" OR "C vulgaris" OR "C japonica" OR rapeseed* OR "B napus" OR raspberr* OR rubus OR "R idaeus" OR blackberr* OR mulberr* OR loganberr* OR safflower* OR carthamus OR "C tinctorius" OR cotton OR gossypium OR serradella* OR birdsfoot* OR ornithopus OR "O sativus" OR sesam* OR "S indicum" OR soybean* OR glycine OR "G max" OR soja OR soya OR "Bay leave*" OR laurus OR "L nobilis" OR dill OR anethum OR "A graveolens" OR fenugreek OR trigonella OR saffron OR crocus OR "C sativus" OR thyme OR thymus OR turmeric OR curcuma OR spinach* OR spinacia OR "S oleracea" OR tetragonia OR "T espansa" OR artiplex OR "A hortensis" OR strawberr* OR fragaria OR "Sugar beet*" OR beta OR "B vulgaris" OR sunflower* OR helianthus OR "H annuus" OR "Sweet potato*" OR ipomoea OR batata* OR "I batatas" OR tangerine* OR mandarin* OR clementine* OR "c reticulata" OR satsuma OR "C unshiu" OR turnip* OR "B rapa" OR vetch* OR vicia OR "V sativa" OR "Viper's grass" OR scorzonera OR "S hispanica" OR walnut* OR juglans OR "J regia" OR watermelon* OR citrullus OR "C vulgaris" ) View Less</p>
1	<p>TITLE-ABS- KEY ( crop OR crops OR flower* OR floral OR fruit* OR *bee OR *bees OR bee OR bees )</p>

### Web of Science. Core Collection

Set	Query
# 6	#5 AND #4 AND #3 Indexes=SCI-EXPANDED, CPCI-S, ESCI, CCR-EXPANDED, IC Timespan=All years
# 5	TS=( sugar OR sugars) NEAR/7 (composition* OR concentration* OR content OR contents OR volume*) Indexes=SCI-EXPANDED, CPCI-S, ESCI, CCR-EXPANDED, IC Timespan=All years
# 4	TS=(nectar OR nectars) Indexes=SCI-EXPANDED, CPCI-S, ESCI, CCR-EXPANDED, IC Timespan=All years
# 3	#2 OR #1 Indexes=SCI-EXPANDED, CPCI-S, ESCI, CCR-EXPANDED, IC Timespan=All years
# 2	TS=(Alfalfa OR Medicago OR "M sativa" OR almond OR almonds OR Prunus OR "P amygdalus" OR "P communis" OR Amygdalus OR "A communis" OR anise OR Pimpinella OR "P anisum" OR badian* OR Illicium OR "I verum" OR carawa* OR Carum OR "C carvi" OR coriander* OR Coriandrum OR "C sativum" OR cumin* OR "C cyminum" OR fennel OR Foeniculum OR Juniperus OR "juniper berr*" OR "J communis" OR apple* OR Malus OR "M pumila" OR "M Sylvestris" OR "P sylvestris" OR "M communis" OR apricot* OR Pyrus OR "P malus" OR "P armeniaca" OR Artichoke* OR Cynara OR "C scolymus" OR asparagus OR "A officianils" OR Avocado* OR Persea OR "P Americana" OR Banana* OR Musa OR "M sapientum" OR "M cavendishii" OR "M nana" OR Barley OR Hordeum OR "H disticum" OR "H vulgare" OR Phaseolus OR blueberr* OR "wild bilberr*" OR whortleberr* OR Vaccinium OR "V myrtillus" OR "V Corymbosum" OR cranberr* OR "myrtle berr*" OR bean OR beans OR faba OR Buckwheat* OR FAGOPYRON OR "F esculentum" OR Cabbage* OR Brassica OR "B oleracea" OR "pak choi" OR pakchoi OR "B chinensis" OR "brussels sprout*" OR collard* OR kale OR kales OR kohlrabi* OR Carob OR carobs OR Ceratonia OR "C siliqua" OR Carrot* OR Daucus OR "D carota" OR "Castor oil seed*" OR "castor oil plant*" OR Ricinus OR "R communis" OR Cauliflower* OR broccoli* OR Cherry OR cherries OR "P avium" OR Cerasus OR "C avium" OR Chestnut* OR Castanea OR "C vesca" OR "C vulgaris" OR "C sativa" OR "Chick pea*" OR chickpea* OR Cicer OR "C arietinum" OR Chicor* OR "C intybus" OR Chilli OR Chillies OR pepper* OR paprika* OR Capsicum OR "C frutescens" OR "C annum" OR allspice* OR Pimenta OR "P officinalis" OR Clover* OR Trifolium OR Coffee* OR Coffea OR "C arabica" OR "C robusta" OR "C liberica" OR "Cow pea*" OR cowpea* OR "blackeye pea*" OR Vigna OR "V unguiculata" OR Cranberr* OR Vaccinium OR "V macrocarpon" OR "Voxycoccus" OR Cucumber* OR gherkin* OR Cucumis OR "c sativus" OR Currant* OR goosecurrant* OR Ribes OR "R nigrum" OR "R rubrum" OR Elder* OR Sambucus OR "S nigra" OR Garlic* OR Allium OR "A sativum" OR Gooseberr* OR "R grossularia" OR Grapefruit* OR pomelo* OR Citrus OR "C maxima" OR "C grandis" OR "C paradisi" OR Grapes OR Vitis OR "V vinifera" OR Bent OR bents OR redtop OR redtops OR "fiorin grass" OR Agrostis OR bluegrass OR poa OR "Columbus grass" OR Sorghum OR "S alnum" OR fescue OR Festuca OR napier OR "elephant grass" OR pennisetum OR "P purpureum" OR "orchad grass" OR Dactylis OR "D glomerata" OR "Rhodes grass" OR Chloris OR "C gayana" OR Hemp OR Cannabis OR "C sativa" OR Leek OR leeks OR "A porrum" OR chive OR chives OR "A schoenoprasum" OR Bridesfoot* OR trefoil* OR lotus OR "L corniculatus" OR lespedeza OR kudzu OR Prueraria OR "P lobata" OR sesbania OR sainfoin OR esparcette OR Onobrychis OR "O sativa" OR sulla OR

Hedysarum OR "H coronarium" OR Lemon\* OR lime\* OR "C limon" OR "C aurantifolia" OR "C limetta" OR Lentil\* OR Lens OR "L esculenta" OR "E lens" OR "L culinaris" OR Linseed\* OR Linum OR "L usitatissimum" OR flaxseed\* OR Lupins OR lupinus OR Melon\* OR Cucumis OR "C melo" OR "Mustard seed\*" OR "White mustard" OR "B alba" OR "B hirta" OR sinapis OR "black mustard" OR "B nigra" OR Okra OR okras OR Abelmoschus OR "A esculentus" OR Hibiscus OR "H esculentus" OR gombo OR Onion\* OR "A cepa" OR Orange\* OR "C sinensis" OR "C aurantium" OR Peach\* OR nectarin\* OR "P persica" OR "A persica" OR persica OR "P laevis" OR Pear\* OR Pyrus OR "P communis" OR Pea OR peas OR Pisum OR "P sativum" OR "P arvense" OR Peppermint OR Mentha OR "M piperita" OR Persimmon\* OR Diospyros OR "D kaki" OR "D virginiana" OR Plum OR plums OR greengage\* OR Mirabelle\* OR damson\* OR "P domestica" OR Sloe\* OR "P spinosa" OR Pumpkin\* OR squash\* OR gourd\* OR Cucurbita OR marrow\* OR Pyrethrum\* OR Chrysanthemum OR "C cinerariifolium" OR Quince\* OR Cydonia OR "C oblonga" OR "C vulgaris" OR "C japonica" OR Rapeseed\* OR "B napus" OR Raspberr\* OR Rubus OR "R idaeus" OR blackberr\* OR mulberr\* OR loganberr\* OR Safflower\* OR Carthamus OR "C tinctorius" OR Cotton OR Gossypium OR Serradella\* OR birdsfoot\* OR Ornithopus OR "O sativus" OR Sesam\* OR "S indicum" OR Soybean\* OR Glycine OR "G max" OR soja OR soya OR "Bay leave\*" OR Laurus OR "L nobilis" OR dill OR Anethum OR "A graveolens" OR fenugreek OR Trigonella OR saffron OR Crocus OR "C sativus" OR thyme OR thymus OR turmeric OR Curcuma OR Spinacia OR "S oleracea" OR Tetragonia OR "T espansa" OR Artiplex OR "A hortensis" OR Strawberr\* OR Fragaria OR "Sugar beet\*" OR Beta OR "B vulgaris" OR Sunflower\* OR Helianthus OR "H annuus" OR "Sweet potato\*" OR Ipomoea OR batata\* OR "I batatas" OR Tangerine\* OR mandarin\* OR clementine\* OR "c reticulata" OR satsuma OR "C unshiu" OR Turnip\* OR "B rapa" OR Vetch\* OR Vicia OR "V sativa" OR "Vipers grass" OR Scorzonera OR "S hispanica" OR Walnut\* OR Juglans OR "J regia" OR Watermelon\* OR Citrullus OR "C vulgaris")Indexes=BCI Timespan=All years

# 1 TS=(( crop OR crops OR flower\* OR floral OR fruit\* OR \*bee OR \*bees OR bee OR bees))  
 Indexes=SCI-EXPANDED, CPCI-S, ESCI, CCR-EXPANDED, IC Timespan=All years

## Zoological Record

Set	Query
# 6	#5 AND #4 AND #3 Indexes=Zoological Record Timespan=All years
# 5	TS=( sugar OR sugars) NEAR/7 (composition* OR concentration* OR content OR contents OR volume*) Indexes=Zoological Record Timespan=All years
# 4	TS=(nectar OR nectars) Indexes=Zoological Record Timespan=All years
# 3	#2 OR #1 Indexes=Zoological Record Timespan=All years
# 2	TS=(Alfalfa OR Medicago OR "M sativa" OR almond OR almonds OR Prunus OR "P amygdalus" OR "P communis" OR Amygdalus OR "A communis" OR anise OR Pimpinella OR "P anisum" OR badian* OR Illicium OR "I verum" OR carawa* OR Carum OR "C carvi" OR coriander* OR Coriandrum OR "C sativum" OR cumin* OR "C cyminum" OR fennel OR Foeniculum OR Juniperus OR "juniper berr*" OR "J communis" OR apple* OR Malus OR "M pumila" OR "M Sylvestris" OR "P sylvestris" OR "M communis" OR apricot* OR Pyrus OR "P malus" OR "P armeniaca" OR Artichoke* OR Cynara OR "C scolymus" OR asparagus OR "A officianils" OR Avocado* OR Persea OR "P Americana" OR Banana* OR Musa OR "M sapientum" OR "M cavendishii" OR "M nana" OR Barley OR Hordeum OR "H disticum" OR "H vulgare" OR Phaseolus OR blueberr* OR "wild bilberr*" OR whortleberr* OR Vaccinium OR "V myrtillus" OR "V Corymbosum" OR cranberr* OR "myrtle berr*" OR bean OR beans OR faba OR Buckwheat* OR Fagopyrum OR Fagopyron OR "F esculentum" OR Cabbage* OR Brassica OR "B oleracea" OR "pak choi" OR pakchoi OR "B chinensis" OR "brussels sprout*" OR collard* OR kale OR kales OR kohlrabi* OR Carob OR carobs OR Ceratonia OR "C siliqua" OR Carrot* OR Daucus OR "D carota" OR "Castor oil seed*" OR "castor oil plant*" OR Ricinus OR "R communis" OR Cauliflower* OR broccoli* OR Cherry OR cherries OR "P avium" OR Cerasus OR "C avium" OR Chestnut* OR Castanea OR "C vesca" OR "C vulgaris" OR "C sativa" OR "Chick pea*" OR chickpea* OR Cicer OR "C arietinum" OR Chicor* OR "C intybus" OR Chilli OR Chillies OR pepper* OR paprika* OR Capsicum OR "C frutescens" OR "C annum" OR allspice* OR Pimenta OR "P officinalis" OR Clover* OR Trifolium OR Coffee* OR Coffea OR "C arabica" OR "C robusta" OR "C liberica" OR "Cow pea*" OR cowpea* OR "blackeye pea*" OR Vigna OR "V unguiculata" OR Cranberr* OR Vaccinium OR "V macrocarpon" OR "Voxycooccus" OR Cucumber* OR gherkin* OR Cucumis OR "c sativus" OR Currant* OR goosecurrant* OR Ribes OR "R nigrum" OR "R rubrum" OR Elder* OR Sambucus OR "S nigra" OR Garlic* OR Allium OR "A sativum" OR Gooseberr* OR "R grossularia" OR Grapefruit* OR pomelo* OR Citrus OR "C maxima" OR "C grandis" OR "C paradisi" OR Grapes OR Vitis OR "V vinifera" OR Bent OR bents OR redtop OR redtops OR "fiorin grass" OR Agrostis OR bluegrass OR poa OR "Columbus grass" OR Sorghum OR "S alnum" OR fescue OR Festuca OR napier OR "elephant grass" OR pennisetum OR "P purpureum" OR "orchad grass" OR Dactylis OR "D glomerata" OR "Rhodes grass" OR Chloris OR "C

gayana" OR Hemp OR Cannabis OR "C sativa" OR Leek OR leeks OR "A porrum" OR chive OR chives OR  
 "A schoenoprasum" OR Birdsfoot\* OR trefoil\* OR lotus OR "L corniculatus" OR lespedeza OR kudzu OR  
 Prueraria OR "P lobata" OR sesbania OR sainfoin OR esparcette OR Onobrychis OR "O sativa" OR sulla OR  
 Hedysarum OR "H coronarium" OR Lemon\* OR lime\* OR "C limon" OR "C aurantifolia" OR "C limetta" OR  
 Lentil\* OR Lens OR "L esculenta" OR "E lens" OR "L culinaris" OR Linseed\* OR Linum OR "L  
 usitatissimum" OR flaxseed\* OR Lupins OR lupinus OR Melon\* OR Cucumis OR "C melo" OR "Mustard  
 seed\*" OR "White mustard" OR "B alba" OR "B hirta" OR sinapis OR "black mustard" OR "B nigra" OR  
 Okra OR okras OR Abelmoschus OR "A esculentus" OR Hibiscus OR "H esculentus" OR gombo OR Onion\*  
 OR "A cepa" OR Orange\* OR "C sinensis" OR "C aurantium" OR Peach\* OR nectarin\* OR "P persica" OR  
 "A persica" OR persica OR "P laevis" OR Pear\* OR Pyrus OR "P communis" OR Pea OR peas OR Pisum OR  
 "P sativum" OR "P arvense" OR Peppermint OR Mentha OR "M piperita" OR Persimmon\* OR Diospyros OR  
 "D kaki" OR "D virginiana" OR Plum OR plums OR greengage\* OR Mirabelle\* OR damson\* OR "P  
 domestica" OR Sloe\* OR "P spinosa" OR Pumpkin\* OR squash\* OR gourd\* OR Cucurbita OR marrow\* OR  
 Pyrethrum\* OR Chrysanthemum OR "C cinerariifolium" OR Quince\* OR Cydonia OR "C oblonga" OR "C  
 vulgaris" OR "C japonica" OR Rapeseed\* OR "B napus" OR Raspberr\* OR Rubus OR "R idaeus" OR  
 blackberr\* OR mulberr\* OR loganberr\* OR Safflower\* OR Carthamus OR "C tinctorius" OR Cotton OR  
 Gossypium OR Serradella\* OR birdsfoot\* OR Ornithopus OR "O sativus" OR Sesam\* OR "S indicum" OR  
 Soybean\* OR Glycine OR "G max" OR soja OR soya OR "Bay leave\*" OR Laurus OR "L nobilis" OR dill OR  
 Anethum OR "A graveolens" OR fenugreek OR Trigonella OR saffron OR Crocus OR "C sativus" OR thyme  
 OR thymus OR turmeric OR Curcuma OR Spinach\* OR Spinacia OR "S oleracea" OR Tetragonia OR "T  
 espansa" OR Artiplex OR "A hortensis" OR Strawberr\* OR Fragaria OR "Sugar beet\*" OR Beta OR "B  
 vulgaris" OR Sunflower\* OR Helianthus OR "H annuus" OR "Sweet potato\*" OR Ipomoea OR batata\* OR  
 "I batatas" OR Tangerine\* OR mandarin\* OR clementine\* OR "c reticulata" OR satsuma OR "C unshiu"  
 OR Turnip\* OR "B rapa" OR Vetch\* OR Vicia OR "V sativa" OR "Vipers grass" OR Scorzonera OR "S  
 hispanica" OR Walnut\* OR Juglans OR "J regia" OR Watermelon\* OR Citrullus OR "C vulgaris")  
 Indexes=Zoological Record Timespan=All years

# 1 TS=(( crop OR crops OR flower\* OR floral OR fruit\* OR \*bee OR \*bees OR bee OR bees))  
 Indexes=Zoological Record Timespan=All years