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Collection and analysis of pesticide residue data for pollen and nectar

Final Report

Benaki Phytopathological Institute (BPI)[1], Agilis S.A- Statistics and Informatics (Agilis)[2]

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Abstract

In the frames of the project "Collection and analysis of pesticide residue data for pollen and nectar", carried out under the contract OC/EFSA/PRAS/2015/08, new available information derived from residue trials were collected, evaluated and used 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 and (if possible) landscape dependent dilution factor. Furthermore, 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 behavior properties of the substances. The detailed screening procedure revealed 125 relevant studies with residues analysis in pollen, nectar and other bee relevant matrices. The relevant data from these studies were captured in an MS Excel database specifically developed for the needs of this project. The identified studies have been conducted in different countries inside and outside Europe, on different crops, while the analysed matrices for pesticide residues were mainly nectar, pollen (collected from bees, hives or directly from the flowers), larvae, wax, honey, flowers and plant parts. Furthermore, RUD values (Residue Unit Doses) were calculated from the measured residues 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 an adequate number of time points after pesticide application, DT_{50} and DT_{90} values for each matrix were calculated. RUD and DT_{50} values were evaluated according to predefined criteria and only the reliable values were further used. Furthermore, in the frame of this project a data analysis was performed in order to investigate potential correlations between residue levels in pollen and nectar; residue levels in pollen or nectar and residue levels in plant foliage; residue levels in pollen or nectar and physicochemical and environmental properties of active substances (i.e. solubility in water, logPow and Koc) and finally residue decline in pollen or nectar and residue decline in other environmental matrices (i.e. soil and water). The analysis revealed that statistically significant differences were detected between residue levels in nectar and pollen, with the residue levels in nectar being lower than the respective levels in pollen. As regards the type of crop, the residue levels in rapeseed notably exceeded the residue levels in *Phacelia tanacetifolia* and other types of crop, both for pollen and nectar. In addition, substance related differences have been detected in RUDs in nectar and pollen, although in some cases the dataset used for data analysis was remarkably confined. A weak positive correlation between the residue levels in nectar with solubility and Koc was observed, while no correlation was detected between the residue levels in pollen with solubility in water and logPow values. Finally, the residue decline values between nectar and pollen were uncorrelated with the respective values in soil and water.

Key words: bees, pesticide residues, pollen, nectar, sugar content

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Summary

The overall objective of the project "Collection and analysis of pesticide residue data for pollen and nectar" (OC/EFSA/PRAS/2015/08) was to collect and evaluate the new available data derived from residue trials conducted from 2010 onwards in order to create a database for pesticide residue levels and residue decline in pollen and nectar, sugar content of nectar, protein content of pollen and where possible landscape dependent dilution factor. In addition, data analysis of the collected data was performed. More specifically, potential correlations between the residue levels and decline in pollen and nectar and the residues levels and decline in different matrices were investigated. Finally, possible correlations between residue levels in pollen and nectar and physicochemical and environmental fate and behavior properties of the active substances were examined as well.

During the first months of the project implementation, a detailed screening of the data submitted and evaluated for the peer-review process under Reg. (EC) 1107/2009 at European level was performed in order to identify the new relevant studies (conducted from 2010 onwards) for pesticides residue data on pollen, nectar and other bee's products. During the screening procedure 314 pesticide active substances were screened (AIR II, AIR III and New Active Substances) and 125 relevant studies were identified. Most of these studies were retrieved from the respective regulatory documents (DARs, RARs, Registration Reports, etc) while others have not yet been submitted for approval of pesticide active substances and plant protection products but they were collected by EFSA from the data owners (e.g. chemical industry) and provided to the contractor.

Since the number of identified studies was much higher than the expected one, a prioritization procedure was carried out during the 1st stage of the project implementation (June 2016 – January 2017) for the selection of the most appropriate forty-five (45) studies to be included in the database. The final decision for the selected studies was taken in collaboration with EFSA. Consequently, all relevant data from each study were extracted and captured in the MS Excel database specifically developed for the needs of this project. Later on, and following the Amendment No1 of the Contract, all identified studies (125) were included in the database. The process followed during the database population was in line with the usage instructions described in the 1st Interim report. Residue data on plant foliage derived from residue trials used for MRL setting have been collected as well.

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 GD on bees (EFSA, 2013). The evaluation was mainly focused on the most important parameters and elements that are considered relevant for this specific project. Certain criteria that were discussed and agreed with EFSA were applied in order to allocate each study into relevant categories according to their fit to the purpose of this project.

Following the population of the database with residue data, RUD values were calculated considering the measured residues 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, DT_{50} and DT_{90} values for each matrix were calculated as well.

The RUD and DT_{50} values derived by reliable studies were further evaluated according to criteria discussed and agreed with EFSA and only some of these values were considered reliable and appropriate in order to be further used for (i) data analysis performed in the frame of this project and (ii) enrichment of the data set of RUD values already available in Appendix F of the EFSA bees GD [EFSA Journal 2013:11(7): 3295] and used for pollinators risk assessment.

The most robust RUDs data set was derived by studies with spray foliar application of pesticides. Many reliable RUD values were calculated for nectar and pollen for several active substances. On the contrary, only a limited number of reliable RUD values were derived by studies with granule application or seed treatment.

As regards the residue decline data, reliable DT_{50} values were calculated for pollen and nectar for several active substances. The calculated DT_{50} values were in most of the cases less than 2 days for

both matrices (pollen and nectar) and only in some cases with FOMC kinetic fit, the DT_{50} value in pollen was estimated to be approximately 4 days. Nevertheless, these values are much lower than the default value of 10 days that already is used according to the EFSA GD [EFSA Journal 2013:11(7): 3295].

Furthermore, in the frame of this project a data analysis was performed in order to investigate potential correlations between: (i) residue levels (RUDs) in pollen or nectar versus residue levels in plant foliage; (ii) residue levels (RUDs) in pollen versus residue levels in nectar; (iii) residue levels (RUDs) in pollen or nectar versus physical-chemical properties of the active substance (S, logPow, Koc) and (iv) residue decline in pollen or nectar versus residue decline in other environmental matrices.

As regards residue levels comparison in pollen and nectar, statistically significant differences were detected between nectar, pollen and plant foliage, with the residue levels in nectar being lower than both pollen and plant foliage, while the highest residue levels were observed in pollen. In addition, statistically significant differences were detected among sampling matrices, with the residue levels in both pollen and nectar being highest when extracted from flower than from bees or traps. Further, strong positive correlation was observed between nectar and pollen, with the relationship being the strongest when the residue is extracted from bees. As regards the type of crop, the residue levels in rapeseed notably exceeded the residue levels in *Phacelia tanacetifolia* and other types of crop, both for pollen and nectar. Furthermore, positive correlation was found between nectar and pollen which is stronger when the type of crop was rapeseed. Statistically significant differences have been identified also between plant foliage and pollen, and plant foliage and nectar. Unfortunately, the possible effect of the direction (downward vs sideward application) of the pesticide application on RUD levels in nectar and pollen was not possible to be studied, because the number of available observations of sideward application was significantly small. In addition, substance related differences have been detected in the residue levels in nectar and pollen. However, in some cases the available data sets were insufficient to draw a clear conclusion.

Regarding investigation of possible correlations between residue levels in nectar and pollen, and physicochemical and environmental properties of the active substance the main focus was on the solubility in water, logPow and Koc value. The results revealed a weak positive correlation between the residue levels in nectar with solubility and Koc values. No correlation has been detected between the residue levels in pollen with solubility in water and logPow values. However, no concrete results could be identified for the residue in nectar and logPow, and the residue in pollen and Koc values.

Finally, possible correlations between residue decline (DT_{50} values) in nectar or pollen with residue decline in soil and water were examined, but the results revealed no correlation of the residue decline values between nectar and pollen with the respective values in soil and water.

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1. Introduction

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

In 2013, EFSA published the "Guidance document on the risk assessment of plant protection products on bees (*Apis mellifera*, Bombus spp. and solitary bees)" to be used under Regulation (EC) 1107/2009. Due to limited availability of data such as residue levels in pollen and nectar, residue decline in pollen and nectar, sugar content of the nectar and protein content of pollen at the time of Guidance Document (GD) development, worst case assumptions were considered resulting in a conservative exposure assessment, especially at lower tiers of the assessment scheme.

The overall objective of the present project was to collect and evaluate the new available information from residue trials conducted from 2010 onwards in order to create a MS Excel database for pesticide residue levels and residue decline in pollen and nectar, sugar content of nectar, protein content of pollen and (if possible) landscape dependent dilution factor. In addition, data analysis of the collected data was performed. More specifically, potential correlations between the residue levels and decline in pollen and nectar and the residues levels and decline in different matrices were investigated. Finally, possible correlations between residue levels in pollen and nectar and physicochemical and environmental fate and behavior properties of the active substances were examined as well.

Within the project the following work packages (WPs) have been defined:

- WP1: Project Management
- WP2: Screening of Assessment Reports, Registration Reports and equivalent documents
- WP3: Development of the Assessment Protocol and Database Structure
- WP4: Database population
- WP5: Data analysis
- WP6: Reporting

According to the Technical Offer and the respective Service Contract the Final Report of this project includes:

- The description of the methods used for the screening of Assessment Reports, Registration Reports and other equivalent documents and for the identification of the relevant studies to be assessed, as described in WP2.
- The results of screening procedure and the complete list with the references found, in WP2.
- The description of the methods used for the assessment of the data as developed under WP3.
- The final database with the collected data according to the objectives 3, 4 and 5 of Tender Specifications.
- The results of data analysis according to the objective 6 of Tender Specifications.
- Overall conclusions

This contract/grant was awarded by EFSA to:

Contractor/Beneficiary: Benaki Phytopathological Institute (BPI)

Contract/Grant title: Collection and analysis of pesticide residue data for pollen and nectar

Contract/Grant number: OC/EFSA/PRAS/2015/08

2. Data and Methodologies

2.1. Screening of regulatory documents for the identification of relevant studies

In the frame of WP2 of the project a detailed screening of the data submitted and evaluated for the peer – review process under Reg. (EC) 1107/2009 at European level was performed in order to identify and list the substances with studies with residue data for pollen and nectar and estimation of sugar content of nectar. Furthermore, studies with residues measurements in other relevant matrices (i.e. flower of the crop, honeydew, guttation fluid, foliage, wax, brood food, etc) were also considered.

The main focus was on the following categories of active substances, since residue studies for pollen and nectar are most likely to have been conducted and submitted for their peer-review process:

- AIR II substances as listed in Reg (EC) No 1141/2010
- AIR III substances as listed in Reg (EC) No 686/2013
- New active substances (NAS) as listed in the EU Pesticide Database (http://ec.europa.eu/sanco_pesticides/public/?event=homepage&language=EN)

For each one of the above mentioned substances the following regulatory documents were screened:

- Draft Assessment Reports (DARs) and Addenda
- (Draft) Renewal Assessment Reports [(d)RARs] and Addenda
- Addenda regarding the evaluation of confirmatory data of active substances
- Registration Reports (RRs) of Plant Protection Products (PPPs) of the aforementioned active substances.

The above mentioned regulatory documents were downloaded from the CIRCA BC (DARs, RARs, Addenda, RRs) and EFSA DMS (DARs, RARs, Addenda) websites. If none of these documents were available a screening in the EFSA conclusion was performed in order to identify any relevant study.

The screening procedure covered the period from 2010 to July 2016 since it is expected that only these new studies will follow the principles described in Appendix G of the EFSA GD on bees (EFSA, 2013).

In addition, other relevant studies which have not yet been submitted for approval of pesticide active substances and plant protection products were collected by EFSA from the data owners (e.g. chemical industry). These studies were provided to the contractor by EFSA during the project's kick-off meeting. The contractor ensures the compliance with existing intellectual property rights and confidentiality rules in relation to these unpublished industry data.

Furthermore, the websites of international organisations and evaluating authorities from outside of Europe (e.g. North and South America) have been searched in order to identify any relevant documents, if publicly available. The screened websites were the following:

- The US EPA website for pesticides (http://www.epa.gov/pesticides) and more specifically the "Protecting Bees and Other Pollinators from Pesticides" area (http://www.epa.gov/pollinator-protection).
- The California EPA (http://www.cdpr.ca.gov/docs/enforce/pollinators/)

- The Health Canada's Pest Management Regulatory Agency (PMRA) (http://www.hc-sc.gc.ca/cps-spc/pest/agri-commerce/pollinators-pollinisateurs/index-eng.php

2.2. Development of the assessment protocol for the evaluation of the identified studies

In the frame of WP3 (Activity 3.1) of the project a detailed assessment protocol has been developed for the evaluation of the identified studies. It is acknowledged that the higher tier studies are carried out in order to assess the exposure of bees to pesticide residues in nectar and pollen under realistic conditions. The assessment protocol was developed based on the principles of Appendices G and S of the EFSA GD on bees (EFSA, 2013) considering also the requirements of the Tender Specifications as incorporated in the Technical Offer and the respective Service Contract.

This protocol describes the key elements / parameters to be considered when evaluating the quality of identified studies with pesticide residue data for pollen and nectar.

The evaluation should focus on the following parameters:

- Experimental conditions
  - Trial conditions (trial identification number, type of higher-tier study, location of the study site, landscape description, plot size, number of replicate test plots, distance between the test plot and the hive, time at which bee colonies were put in position, *etc.*)
  - Crop related parameters (e.g. crop and crop variety, seasonal growth cycle of crop, calendar date and time of the start and the end of the flowering period of the crop, crop density, *etc.*)
  - Test organisms (e.g. bee species, colony size, health status, bee behaviour including foraging activity before exposure and during the study, *etc.*)
- Application of treatment
  - Identity of test item (e.g. name, batch/lot number, type of formulated product, identity and content of the containing active ingredient, *etc.*)
  - Application methodology (e.g. foliar spraying, seed treatment, granule application)
  - Application equipment used
  - Application pattern (e.g. application rate, number of applications, application interval)
  - Direction of application (in case of spraying)
  - Dust deposition onto bare soil (in case of seed treatment and granule application)
  - Time of pesticide application (including calendar date and time during the day, i.e. morning, daytime, evening)
  - Growth stage of the crop at the time of application (BBCH stage)
  - Time between the pesticide application and the start of blooming
  - Meteorological data / weather conditions at the time of application (temperature, air humidity, wind speed, rainfall, *etc.*)
- Sampling
  - Type, matrix and number of samples (nectar directly from plants, nectar in honey sacks from bees actively foraging in the treated crop or bees returning to hives, pollen samples

directly from plants, pollen from bees, pollen from pollen traps, other samples such as flower samples, hive samples, aphid (or other insects) honeydew, guttation fluid, *etc.*)

- Spatial variation (type of sampling area e.g. treated field, crop at field margins, adjacent crops, number of sampling locations, position of sampling locations, *etc.*)
- Temporal variation (time between pesticide application and sampling, distribution of sampling time points over the flowering period of the crop, time of sampling during the sampling day, time at which sampling started in relation to the growing season of the crop, *etc.*)
- Sampling methodology (methodology used for the collection of each sample type, conditions during transportation of collected samples to the test facility, conditions during storage at the laboratory until analysis, *etc.*)

- Analysis of collected samples
  - Analytical methodology (detailed description of all analytical methods used including reporting of their sensitivity and validation parameters, *etc.*)
  - Analysis of collected samples (concentration of each analyte for each trial – sampling time – sample matrix combination, statistical methods used for raw data analysis, *etc.*)

The detailed protocol for the assessment and evaluation of the identified studies is presented in Appendix – A.

2.3. Development of the database structure

The structure of the Excel database stemmed from a thorough data requirement analysis. Defining and analysing the user requirements, constituted the first step undertaken for the development of the Excel database. Namely, that analysis was carried out with the view to thoroughly specify the needs that the Excel database will serve, the structure of the Excel database and the organization of the information, and the functionalities that the database would offer to its end users.

The twofold requirements that the Excel database serves include the (a) storage of the essential information extracted from the relevant studies and other documentation (data entry process) and (b) (re)view of the information stored. Following the above-mentioned requirements, the Excel database was structured in a way to facilitate its users to entry the information but also to enable them (re)view the recorded data in their entirety.

For the needs of the data requirement analysis, we have identified (a) the essential information to be recorded under each column (a.k.a. 'fields'), (b) the main entities and (c) the logical the relationships among the identified entities.

The conceptual framework based on which the database was structured follows the logical relationships among the identified entities. As depicted in Figure 110, the main entities identified are the following: (1) the study, (2) the trial, (3) the treatment group, (4) the sampling, (5) application, (6) the test item and (6) the active substance (depicted in Figure 1). The identified entities and their relationships are described in the following paragraphs.

The "study" is the main entity of the conceptual model representing an actual study conducted for measuring residue data in pollen and nectar. For each study several metadata are recorded, such as year, title and authors.

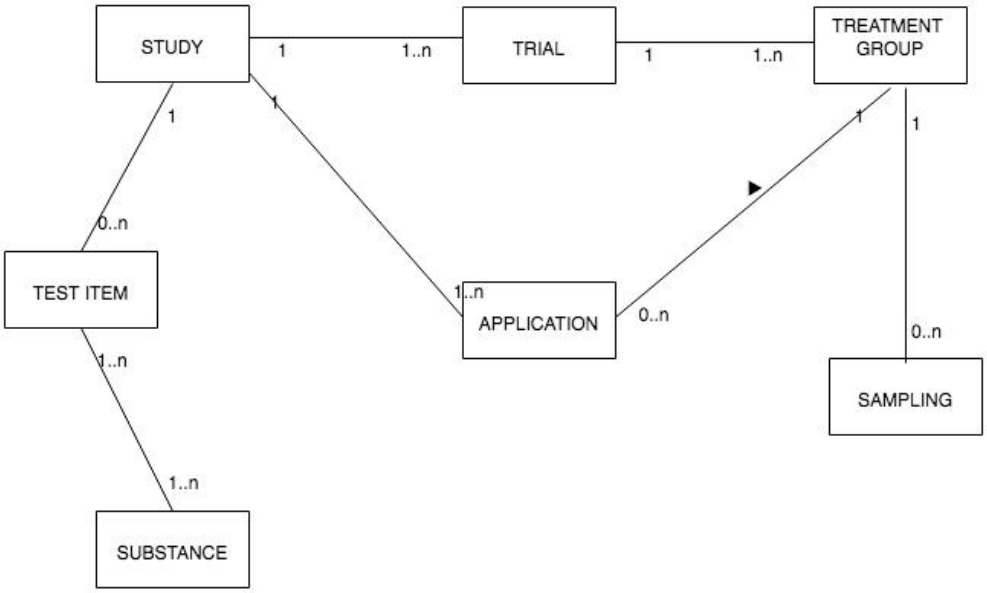


Figure 1: High-level conceptual model of the Excel database.

A "study" uses one pesticide formulation (plant protection product), which is represented by the "test item" entity. The latter, in turn, is composed of one or more active substances and/or metabolites, which are both classified under the "substances" entity in the information model.

Under the framework of each study, one or more trials may be undertaken in a particular geographical area, whereas for each trial usually two treatment groups are defined; the treated and the control. The treatment groups store information about the crop (i.e. crop name, crop type, etc.) and the location. It should be noted that in the implemented Excel database the "Trial" and "Treatment Group" entities have been merged into one.

The one-to-many relationship between "treatment group" and the "application" entities indicates that a "test item" is applied to a treatment group many times. Indicatively the "application" and the "seed treatment" include information about the type of application, the application date, method, equipment etc.

Finally, for each "treatment group" many samples are taken in order to measure the residue levels and in some cases the sugar and protein content. These measures are represented by the "sampling" entity. Moreover, when the sampling takes place after the application, the sampling is related to a specific application.

The Excel database was thus organised in separate Excel worksheets following the principles of the conceptual model, minimising data redundancy, with the view improve data integrity and facilitate the data entry process. Thus, the Excel database follows a structure that prevents the end user from recording the same information in more than one worksheets.

The Excel database is composed of **nine Excel worksheets**:

- Sheet '*DROPDOWNS*': contains the lists used for database population.
- Sheet '*SUBSTANCES*': contains information about the measured/analysed substances.
- Sheet '*STUDIES*': contains information about the study as such.

- Sheet '*TREATMENT_GROUPS*': contains information about the trials and the treatment groups defined in each study.
- Sheet '*APPLICATIONS*': contains information about the applications performed in each study (i.e. spray application, granule application, drench application, drip irrigation)
- Sheet '*SEED_TREATMENTS*: contains information about the seed treatments performed in each study.
- Sheet '*SAMPLINGS*': contains information about the samplings and the calculated residue levels.
- Sheet '*CALCUCATED_ DT_{50}_DT_{90}*': contains the calculated DT_{50} and DT_{90} values.
- Sheet '*FOLIAGE_RUD*': contains the residues levels (and the respective RUD values) on plant foliage.
- Sheet '*ALL_DATA_MERGED*: combines all information recorded in the sheets '*STUDIES*', '*TREATMENT GROUPS*', '*APPLICATIONS*', and '*SAMPLINGS*'.

All but the last sheet, are used for the data entry process, while the last one, namely, the sheet '*ALL_DATA_MERGED*' facilitates the viewing of the data, bringing together the data stored for all studies.

At the initial stage of drafting of database structure a pilot phase of database population was carried for which the whole procedure was applied for five (5) selected studies. During the pilot phase of data entry a communication with EFSA was carried out in order to discuss the functionality of the database. The received comments were considered for the preparation of the final version of database.

2.4. Database population process

The process followed during the database population was in line with the usage instructions described in the 1st Interim report (Section 3.4.2. Data entry process – usage instructions).

More specifically, for each one of the selected studies the steps described below were followed for the data entry:

Step 1: "*DROPDOWNS*" worksheet population

Firstly, the sheet '*DROPDOWNS*' was developed. For each field/column populated in the finally structured database (e.g. country name, crop name, crop type, *etc.*), all allowable values were listed in the respective "dropdown" list in an effort to have a harmonized description of all parameters entered. However, since it was not possible to have foreseen all possible inputs for each field beforehand when structuring the database, there were cases where the 'value' to be entered was not already in the dropdown list. Then the user had first to add the new specific 'value' at the end of the respective list in the sheet '*DROPDOWNS*'. For instance, if the user needed to include a new value for a test species, this was entered at the end of the list "*Test Species*" (Column P) of this sheet and then the user could select this value to be entered in a subsequent sheet.

This sheet contains the following lists:

Column 'Country Name': List of countries. The list of the countries used follows the standard ISO 2-digit codes.

Column 'Crop Name': List of the main agricultural crops in Europe, potentially used in the selected studies.

Column 'Crop Type': List of the main crop types (e.g. annual crop, perennial crop, succeeding crop, etc.).

Column 'Application Method': List of the relevant forms of pesticide applications (e.g. spraying, seed treatment, granule application).

Column 'Sampling Matrix': List with sample types (sampling matrices) e.g. honey from comb/hive, nectar from bees, nectar from combs, plant parts etc.

Column 'Unit Name': List of all units of measurement (e.g. Gram/100 gram, Microgram/kilogram, hectares) that were considered to be relevant for this project.

Column 'Unit Short Name': List of all units (short name) of measurement (e.g. g/100g, µg/kg, ha) that are considered to be relevant for this project.

Column 'Type of residue level measure': List of the type of residue level measure with values average, maximum, minimum, individual residue values per sample, etc. The option "not mentioned" is relevant when this information was not provided in the study report.

Column 'Test Species': List with different species of bees e.g. bumble bees, honeybee, solitary bees, etc. The option "None" is relevant when no bees have been used in the study.

Column 'Formulation Type': List of common types of pesticide formulations (EC, SC, WP, etc.). The list was used in the sheet "Studies" to define the formulation type of the test item.

Column 'Time of Day': List with the most important times or parts of the day. It was used in sheets 'Applications' and 'Samplings' to describe the exact time of the day the application of the test item or the sampling was carried out.

Column 'Type of higher tier study": List of the different types of higher tier study (e.g. field, tunnel, glasshouse).

Column 'Answers: List of possible answers in many fields of the following sheets.

Column 'Visual Assessment": List of possible answers in the visual assessment used in "*CALCULATED_DT_{50}_DT_{90}*" sheet (e.g. excellent, very good, acceptable, etc).

In order to follow a harmonised terminology, some of the used lists were obtained from definitions of the 'Matrix catalogue'[1], if available.

Step 2: "*SUBSTANCES*" worksheet population

Following the population of the worksheet "*DROPDOWNS*", the sheet *'SUBSTANCES*" was filled-in.

This worksheet contains information about the active substance(s), the test item used in the relevant studies, the concentration (nominal/analysed) of the active substance(s) in each test item, *etc*. Furthermore, this sheet contains information related to the physicochemical and environmental

[1] EFSA Matrix Catalogue: http://www.efsa.europa.eu/it/efsajournal/doc/SSD_CAT_MTX.xls

properties of each substance. The relevant data for the metabolites of each active substance (if analysed in the relevant study) were also inserted in this sheet.

During the population process of this sheet, special attention was given when filling the fields "Substance CAS Number" and "Test Item". These fields were never left blank, since they are identifiers for linking together information across the different worksheets. In detail, "Test Item" was used as a means for further linking the active substance with study of reference. Therefore, each active substance was accompanied by a unique identifier ("Substance CAS Number"). The same also hold for the column "Test Item" since for each study one test item has been used.

More specifically, the following data have been included in the sheet "*SUBSTANCES*".

Table 1: Data included in the relevant fields of the sheet "*SUBSTANCES*"

Columns	Description	Type
Substance CAS Number	Substance CAS Number. This field was never left blank, since it was used as identifier for linking together information across the different worksheets. If a metabolite did not have a CAS number, a unique code was defined by the user for this metabolite.	Text
Substance Code	Substance identification code (if available).	Text
Substance Name	Name of the substance.	Text
Is Metabolite?	It indicates if the substance is a metabolite. (Options: Yes, No).	List
Parent Substance CAS Number	It indicates the CAS number of the parent substance for each metabolite.	List
Test Item	Trade name or manufacturers' code of the pesticide formulation in which the active substance was included. This field was never left blank, since it was used as identifier for linking together information across the different worksheets.	Text
Content of a.s. (nominal)	The nominal content of active substance in the test item.	Number
Content of a.s. (analysed)	The analysed content of active substance in the test item, as indicated in the study report.	Number
Content of a.s. (unit)	Records the unit in which the active substance content was expressed.	List
Solubility in water – value	Solubility in water as indicated in the relevant regulatory documents (e.g. EFSA conclusion).	Number
Solubility in water – unit	Records the unit of solubility in water (e.g. mg/L).	List
Solubility in water – pH	Records the pH at which the water solubility was determined.	Number
Log Pow	Log Pow as indicated in the relevant regulatory documents (e.g. EFSA conclusion).	Number

Columns	Description	Type
pH	Records the pH at which the Log Pow was determined.	Number
Soil – DT_{50}	Records the normalised (at 20^0C, pF2 soil moisture) laboratory DT_{50} values in soil as indicated in the relevant regulatory documents (e.g. EFSA conclusion) where available. In case no normalised values were obtained, the reported DT_{50} values from laboratory or field studies have been included in the database. Where an average value (geometric/arithmetic mean) was calculated, it was also included in the database.	Number
Soil – DT_{90}	Records the DT_{50} values in soil as indicated in the relevant regulatory documents (e.g. EFSA conclusion). Please note that in most of the cases no normalised DT_{90} values were available. Where an average value (geometric/arithmetic mean) was calculated, it was also included in the database.	Number
Soil – unit	Records the unit of DT_{50} and DT_{90} (e.g. days).	List
Soil – pH	Records the soil pH at which the DT_{50} and DT_{90} values were determined.	Number
Soil – T (°C)	Records the soil temperature at which the DT_{50} and DT_{90} values were determined.	Number
Soil Type	Records the soil type for which the DT_{50} and DT_{90} values were determined.	Text
Soil – Kinetics	Records the kinetics used for the calculation on DT_{50} and DT_{90} values (e.g. SFO, DFOP, FOMC).	Text
Water/sediment system	Records the water/sediment system as indicated in the relevant regulatory documents (e.g. EFSA conclusion).	Text
Whole system – DT_{50}	Records the DT_{50} values in the whole system as indicated in the relevant regulatory documents (e.g. EFSA conclusion) Where an average value (geometric/arithmetic mean) was calculated, it was also included in the database.	Number
Whole system – DT_{90}	Records the DT_{90} values in the whole system as indicated in the relevant regulatory documents (e.g. EFSA conclusion).	Number
Whole system – unit	Records the unit of DT_{50} and DT_{90} (e.g. days).	List
Whole system - Kinetics	Records the kinetics used for the calculation on DT_{50} and DT_{90} values (e.g. SFO, DFOP, FOMC).	Text
Water – DT_{50}	Records the DT_{50} values in water as indicated in the relevant regulatory documents (e.g. EFSA conclusion) Where an average value (geometric/arithmetic mean) was calculated, it was also included in the database.	Number
Water – DT_{90}	Records the DT_{90} values in water as indicated in the relevant regulatory documents (e.g. EFSA conclusion) Where an average value (geometric/arithmetic mean) was calculated, it was also included in the database.	Number
Water – unit	Records the unit of DT_{50} and DT_{90} (e.g. days).	List
Water – pH	Records the water pH at which the DT_{50} and DT_{90} values were determined.	Number
Water - Kinetics	Records the kinetics used for the calculation on DT_{50} and DT_{90} values (e.g. SFO, DFOP, FOMC).	Text

Columns	Description	Type
Sediment – DT_{50}	Records the DT_{50} values in sediment as indicated in the relevant regulatory documents (e.g. EFSA conclusion) Where an average value (geometric/arithmetic mean) was calculated, it was also included in the database.	Number
Sediment – DT_{90}	Records the DT_{90} values in sediment as indicated in the relevant regulatory documents (e.g. EFSA conclusion) Where an average value (geometric/arithmetic mean) was calculated, it was also included in the database.	Number
Sediment – unit	Records the unit of DT_{50} and DT_{90} (e.g. days)	List
Sediment – pH	Records the sediment pH at which the DT_{50} and DT_{90} values were determined.	Number
Sediment - Kinetics	Records the kinetics used for the calculation on DT_{50} and DT_{90} values (e.g. SFO, DFOP, FOMC).	Text
Hydrolysis –degradation	Records whether hydrolysis degradation occurs for the substance under concern (Options: stable, non-stable).	List
Hydrolysis DT_{50}	Records the DT_{50} value for hydrolysis (relevant only for non-stable substances).	Number
Hydrolysis DT_{50}-unit	Records the unit of DT_{50} (e.g. days).	List
Hydrolysis pH	Records the pH at which the DT_{50} values was determined.	Number
Hydrolysis T (°C)	Records the temperature at which the hydrolysis DT_{50} values were determined.	Number
Soil photolysis	Records whether soil photolysis occurs for the substance under concern (Options: Yes, No).	List
Soil photolysis DT_{50}	Records the DT_{50} value (if soil photolysis occurs).	Number
Soil photolysis DT_{50}-unit	Records the unit of DT_{50} (e.g. days).	List
Readily biodegradable	Indicates whether the substance is readily biodegradable or not (Options: Yes, No).	List
Koc - Value	Koc value as indicated in the relevant regulatory documents (e.g. EFSA conclusion). Arithmetic mean Koc value was presented in the data base where possible.	Number
Koc – pH dependence	Indicates whether the Koc is pH dependent or not (Options: Yes, No).	List
Koc-pH	Records the pH at which adsorption values were determined (relevant only for pH dependent Koc).	Number
Notes	Additional or summarized remarks about the substance that cannot be given under any of the previous columns.	Text

Step 3: "*STUDIES*" worksheet population

The sheet "*STUDIES*" was filled-in with study specific information, such as the title of the study, the authors, the identification code and indication whether the study was GLP compliant. Furthermore, the worksheet contains the trade name or manufactures' code of the pesticide formulation (Test Item) applied and the bee species examined (if any).

During the population process of this sheet, special attention was given when filling the field "Study ID", since a unique identification code should be given for each study under this field permitting the linking of information among the different sheets.

More specifically, the following data have been included in the sheet "*STUDIES*".

Table 2: Data included in the relevant fields of the sheet "*STUDIES*"

Columns	Description	Type
Study ID	The unique identification code of the study, as indicated in the list of relevant studies	Text
Study owner reference number	Study owner reference number as indicated in the list of relevant studies. For certain studies no such reference number is available.	Text
Title	Title of the study.	Text
Author(s)	Author(s) of the study. In cases where the names of the authors were not mentioned in the study report, the phrase "not mentioned" was captured in the database.	Text
Year	Year of study report.	Number
Test Species (Bees)	The bee species used in the study. The selection "none" has been used for studies without bees.	List
Test Item	The pesticide formulation used in the study. Each test item was selected from a dropdown list linked with the "SUBSTANCES" sheet and it was specific for each study. In some cases one test item was used in more than one study.	List
Batch No. of Test Item	The batch number of the pesticide formulation (test item)	Text
Formulation Type of Test Item	The formulation type of the pesticide (Options: WG, EC, SC, WP, OD, etc.)	List
GLP compliance	It indicates if the study was GLP compliant or not. (Options: Yes, No)	List
Number of Trials	Total number of trials defined in study. In some studies different applications have been conducted in different plots or tunnels and the analysed residues were presented separately for each plot/tunnel. In such cases, each plot/tunnel was considered as separate "trial", although it is not correct with the strict sense of the word "trial".	Number
Number of Applications	Total number of applications performed in the study.	Number
Type of higher tier study	Type of higher tier study (Options: Field, Tunnel, Glasshouse)	List
Notes	Additional or summarized remarks about the study that cannot be given	Text

Columns	Description	Type
	under any of the previous columns.	

Step 4: "*TREATMENT_GROUPS*" worksheet population

The sheet "*TREATMENT_GROUPS*" was filled-in with data that concern each specific study.

This worksheet contains information about the treatment groups (Control and Treated Fields) defined in each study's trial. In most studies, two treatment groups were set up per trial, 'Control' group that remained untreated and 'Treated' group that was treated with the test item of the study. For each treatment group, information about the exact location, the country, the area and additional information about the cultivated crops, the flowering period, the landscape description and the history of the field, *etc* was retained. Lastly, the number of applications performed per field was stored in this sheet.

More specifically, the following data were included in the sheet "*TREATMENT_GROUPS*".

Table 3: Data included in the relevant fields of the sheet "*TREATMENT_GROUPS*"

Columns	Description	Type
Study ID	Identification code of the study, used in the sheet "*STUDIES*". The column Study ID is a dropdown list that contains the identification codes of the studies recorded in the "*STUDIES*" sheet.	List
Trial ID	Identification code of the trial. The trial ID is an identification code of the trial and must always have a value. If the study did not provide codes for its trials, these codes were assigned by the user (e.g. Trial-01). In some studies different applications were conducted in different plots or tunnels and the analysed residue values were presented separately for each plot/tunnel. In such cases, each plot/tunnel was considered as separate "trial" (although it is not correct with the strict sense of the word "trial") or the number of the plot/tunnel was added in this column (e.g. Trial-01_Tunnel-e, Trial-01-Plot-T1).	Text
Treatment Group	In general, two treatment groups were set up per trial: 'Control' and 'Treated'. (Options: Control, Treated) The "Treated" field was treated with the test item examined in the study and the control was left untreated or treated with tap water. In some studies there were no control groups (plots or tunnels), but some samples were collected before the application of the test item and considered as control samples.	List
Location	The exact location of the treatment group as indicated in the study report.	Text
Region	The region of the treatment group as indicated in the study report.	Text
Country	Country where the treatment group is located.	List
Intra/Extra-EU	It indicates whether the country is a member state of the European Union (EU). (Options: Intra-EU, Extra-EU)	List

Columns	Description	Type
Area (ha)	Area of treatment group, expressed in hectares. For tunnel studies the area indicates the area of the tunnel.	Number
Crop Name	Name of crop grown in the treatment group. (Options: Alfalfa, Apples, Apricots, Avocados, etc.). In order to follow a harmonised terminology, the crops names used were obtained from definitions of the 'Matrix catalogue'[2].	List
Crop Variety	The crop variety cultivated in the specific treatment group.	Text
Crop Type	Describes the type of crop grown in the treatment group. (Options: annual crop, perennial crop, succeeding crop)	List
Flowering start period	Defines when the flowering period of the crop starts. In most studies this information was not included in the study report.	Date
Flowering end period	Defines when the flowering period of the crop ends. In most studies this information was not included in the study report.	Date
Crop Density	Crop density of the treatment group.	Number
Crop Density (unit)	The unit in which the crop density of the treatment group was measured.	List
Density of flowering weeds in the field	Defines the density of flowering weeds in the field, if this information was included in the study report. For tunnel studies the phrase "not relevant" was used.	Number
Density of flowering weeds in the field (unit)	The unit in which the density of flowering weeds in the treatment group is measured.	List
Distance between the field and the hive (m)	The distance between the hive and the treatment group, expressed in meters. For studies where the hive was located inside the field/tunnel the selection "0" was chosen.	Number
Treatment Group Type	Type of treatment group (Options: Field, Tunnel, Glasshouse).	List
Number of Applications (per treatment group)	Contains the number of times the test item was applied in the specific treatment group. This field was also completed for control groups in case of application with tab water. Note that applications referring to control groups are not stored in the sheet "Applications".	Number
Notes	Additional remarks about the treatment groups and the study that cannot be given under any of the previous columns.	Text
Landscape Description	A brief description of the landscape, if available in the study report.	Text
History of the Field	The history of the field (e.g. crops, pesticide treatments, etc.)	Text

[2] EFSA Matrix Catalogue: http://www.efsa.europa.eu/it/efsajournal/doc/SSD_CAT_MTX.xls

Step 5: "*APPLICATIONS*" worksheet population

The sheet "*APPLICATIONS*" was filled-in with relevant data from each specific study.

The sheet *'APPLICATIONS'* contains information about the applications of the test item (pesticide formulation) performed in each trial. The application method and equipment used, as well as the application rate of the test item and its measured/analysed substances was stored in this sheet.

More specifically, the following data were included in the sheet "*'APPLICATIONS'*".

Table 4: Data included in the relevant fields of the sheet "*APPLICATIONS*"

Columns	Description	Type
Application ID	Identification code of the application. The application ID uniquely identifies the application and must always has a value. These codes were assigned by the user (e.g. Application-1, Application -2, etc.)	Text
Study ID	Identification code of the study. The Study ID is a dropdown list that contains the identification codes of studies recorded in "STUDY" sheet.	Text
Trial ID	Identification code of the Trial. Dropdown list that takes values from the trials recorded in the sheet "*TREATMENT_GROUPS*". The field "Trial ID" takes values only when the user selects a study. More specifically, when a study was selected from the dropdown list (column Study ID) the field Trial ID was populated with the trials assigned in the study.	List
Treatment Group	The treatment group in which the test item was applied. It always took the value "Treated", since applications were carried out only in treated (not control) groups. The applications conducted with tap water in some control groups were not captured in this sheet.	List
Application Date	Records the calendar date of application.	Text
Application method	Records the application method used: spraying, seed treatment, granule application etc.	List
Application equipment	Records the equipment used in the application.	Text
Direction of application (in case of spraying)	Records the direction of the application during spraying (Options: Upward, Downward, Sideward).	List
Dust deposition rate (in case of 'seed treatment' and 'granule application') (mg/ha)	The dust deposition rate during the application, expressed in mg/ha. It was filled-in only when the application method was 'seed treatment' or 'granule application'.	Number
Applications interval (days)	Records the interval (in days) between different applications in the same study. For studies with one application the phrase "not relevant" was used.	Number
Application during active foraging	Indicates if the application took place during active foraging period (Options: Yes, No, Not relevant, Not mentioned). The option "not relevant" was chosen for studies without bees.	List
Time of application	Records the time of application during the day (Options: Morning, Evening, Night, etc.)	List

Columns	Description	Type
Application rate (Test Item)	Records the rate of the pesticide formulation (test item) applied.	Number
Application rate unit (Test Item)	Records the unit in which the application rate was expressed.	List
Substance ID	Records the substance included in the test item used in the application. In this field the options displayed are the identification codes of the substances stored in the sheet '*SUBSTANCES*'. The field "Substance ID" takes values only when the user selects a study. More specifically, when a study is selected from the dropdown list (column Study ID) the field Substance ID is populated with the substances assigned in the study. When the application was conducted with test item containing more than one active substance, the record for the same application was repeated for each active substance. The "Substance ID" field can be populated with values only when the user has selected a study ID (Column B). The metabolites were not included in this field, since the term "application" is relevant only for active substance contained in the applied test item. However, since residues data for metabolites were available in some studies, these data have been captured in "*SAMPLING*" sheet, linked with a specific study and a specific test item, but were not linked to a specific application (see also the relative description in "*SAMPLING*" sheet).	List
Application rate (substance)	The application rate of each active substance. When the application was conducted with test item containing more than one active substance, the record for the same application was repeated for each active substance.	Number
Application rate unit (substance)	The Unit in which the application rate of substance is measured.	List
Growth stage of the crop at the time of application (BBCH Low)	Records the growth stage of the crop at the time of application – lower bound.	Number
Growth stage of the crop at the time of application (BBCH High)	Records the growth stage of the crop at the time of application – upper bound.	Number
Time between the pesticide application and the start of the flowering (days)	Records the time between the pesticide application and the flowering, expressed in days. If the application was conducted during flowering the time between the pesticide application and the start of flowering was set at "0".	Number
Rainfall (l/m2/day)	Records the weather conditions (rainfall) during application. Rainfall measurement expressed in l/m2.	Number
Minimum relative air humidity (%)	Minimum relative air humidity just before or after the application (%)	Number
Maximum relative air humidity (%)	Maximum relative air humidity just before or after the application (%)	Number
Mean relative air humidity (%)	Mean relative air humidity during the application (%)	Number

Columns	Description	Type
Wind speed (m/s)	Wind speed during the application, expressed in m/s.	Number
Minimum Temperature (° C)	Minimum temperature just before or after the application, expressed in ° C.	Number
Maximum Temperature (° C)	Maximum temperature just before or after the application, expressed in ° C.	Number
Mean Temperature (° C)	Mean temperature just before or after the application, expressed in ° C.	Number
Notes	Records any other remarks as regards the application.	Text

Step 5: "*SEED_TREATMENT*" worksheet population

The sheet "*SEED_TREATMENT*" was filled-in with relevant data from each specific study.

The sheet "*SEED_TREATMENT*" contains information about the seed treatment performed in each trial. The equipment used, as well as the application rate and the seed loading/ dressing rate of the measured/analysed substances was stored in this sheet.

More specifically, the following data were included in the sheet "*SEED_TREATMENT*".

Table 5: Data included in the relevant fields of the sheet "*SEED_TREATMENT*"

Columns	Description	Type
Seed Treatment ID	Identification code of the seed treatment. The seed treatment ID uniquely identifies the performed seed treatment and must always has a value. These codes were assigned by the user (e.g. Treatment-1, Treatment -2, etc.)	Text
Study ID	Identification code of the study. The Study ID is a dropdown list that contains the identification codes of studies recorded in "STUDY" sheet.	Text
Trial ID	Identification code of the Trial. Dropdown list that takes values from the trials recorded in the sheet "*TREATMENT_GROUPS*". The field "Trial ID" takes values only when the user selects a study. More specifically, when a study was selected from the dropdown list (column Study ID) the field Trial ID was populated with the trials assigned in the study.	List
Treatment Group	The treatment group in which the test item was applied. It always took the value "Treated", since applications were carried out only in treated (not control) groups.	List
Date of sowing	Records the calendar date of sowing.	Text
Type of sowing machine	Records the type of the sowing machine used.	Text
Dust deposition rate (in case of 'seed treatment' and 'granule application') (mg/ha)	The dust deposition rate during the application, expressed in mg/ha. It was filled-in only when the application method was 'seed treatment' or 'granule application'.	Number
Sowing interval (days)	Records the interval (in days) between different sowings in the same study.	Number

Columns	Description	Type
	In all captured studies only one sowing was performed, therefore the phrase "not relevant" was captured in this cell.	
Sowing during active foraging	Indicates if the sowing took place during active foraging period (Options: Yes, No, Not relevant, Not mentioned). The option "not relevant" was chosen for studies without bees.	List
Time of sowing	Records the time of sowing during the day (Options: Morning, Evening, Night, etc.)	List
Time between the sowing and the start of the flowering	Records the time between the sowing and the flowering, expressed in days.	Number
Application rate (Test Item)	Records the rate of the pesticide formulation (test item) applied.	Number
Application rate unit (Test Item)	Records the unit in which the application rate was expressed.	List
Substance ID	Records the substance included in the test item used in the application. In this field the options displayed are the identification codes of the substances stored in the sheet '*SUBSTANCES*'. The field "Substance ID" takes values only when the user selects a study. More specifically, when a study is selected from the dropdown list (column Study ID) the field Substance ID is populated with the substances assigned in the study. The "Substance ID" field can be populated with values only when the user has selected a study ID (Column B). The metabolites were not included in this field, since the term "application" is relevant only for active substance contained in the applied test item. However, since residues data for metabolites were available in some studies, these data have been captured in "*SAMPLING*" sheet, linked with a specific study and a specific test item, but were not linked to a specific application (see also the relative description in "*SAMPLING*" sheet).	List
Application rate (substance)	The application rate of each active substance.	Number
Application rate unit (substance)	The Unit in which the application rate of substance is measured.	List
Seed loading / dressing rate (substance)	The seed loading / dressing rate of each active substance. In some studies, the seed dressing/loading rate was not included in the study report, therefore it was calculated considering the application rate (kg a.s./ha) and the seeding rate (seeds / ha). In addition, in some cases the available data were not adequate for the calculation of the seed loading / dressing rate used in the study.	Number
Seed loading / dressing rate unit (substance)	The Unit in which the seed loading / dressing rate of substance is measured.	List
Rainfall (l/m2/day)	Records the weather conditions (rainfall) during sowing. Rainfall measurement expressed in l/m2.	Number
Minimum relative air humidity (%)	Minimum relative air humidity just before or after the sowing (%)	Number

Columns	Description	Type
Maximum relative air humidity (%)	Maximum relative air humidity just before or after the sowing (%)	Number
Mean relative air humidity (%)	Mean relative air humidity during the sowing (%)	Number
Wind speed (m/s)	Wind speed during the sowing, expressed in m/s.	Number
Minimum Temperature (° C)	Minimum temperature just before or after the sowing, expressed in ° C.	Number
Maximum Temperature (° C)	Maximum temperature just before or after the sowing, expressed in ° C.	Number
Mean Temperature (° C)	Mean temperature just before or after the sowing, expressed in ° C.	Number
Notes	Records any other remarks as regards the sowing.	Text

Step 6: "*SAMPLINGS*" worksheet population

The sheet "*SAMPLINGS*" was filled-in with data that concern each specific study.

The samples collected before and after application, as well as the sampling matrix used and the methodology followed were recorded in this worksheet. Furthermore, the sheet contains data that stemmed from the analysis of the sample, such as residue levels of each analyte, sugar/protein content determined in the sample, *etc*.

More specifically, the following data were included in the sheet "*SAMPLINGS*".

Table 6: Data included in the relevant fields of the sheet "*SAMPLINGS*"

Columns	Description	Type
Sampling ID	Identification code of the sample taken. The field "Sampling ID" is an identification code of each sample and must always has a unique value. These codes assigned by the user (e.g. 1, 2, 3, etc.) or copied from the study report (if available).	Text
Study ID	Identification code of the study. The column "Study ID" is a dropdown list that contains the identification codes of the studies recorded in "STUDY" sheet.	List
Trial ID	Identification code of the trial (region). The field Trial ID takes values only when the user selects a study. More specifically, when a study is selected from the dropdown list (column Study ID) the field Trial ID is populated with the trials assigned in the study.	List
Treatment Group	It defines if the sample was taken from the control or the treated field. (Options: Control, Treated). Some studies did not comprise "control" group, but sampling was conducted in treated groups before the pesticide application. If these samples were indicated as "control" samples in the study report, they were captured as "control" samples in the database as well.	List
Sampling date	Records the calendar date of sampling.	Date

Columns	Description	Type
Time of sampling	Records the time of sampling during the day (Options: Morning, Evening, Night, etc.).	List
Sampling Matrix	Type of sample taken. (Options: Nectar from bees, Pollen from bees, Nectar from flowers, Pollen from flowers, Honey from hive, Plant parts, Wax, Guttation fluid, Brood food, Honey from combs, etc.)	List
Sampling Methodology	Records a brief description of the sampling methodology used, as retrieved from the study report.	Text
Deviations from the recommendations of EFSA GD	Records the main deviations of the study from the recommendations of EFSA GD and the assessment protocol developed under Activity 3.1 of WP3. More details as regards the study evaluation are presented in the respective "Evaluation check list" (Appendix E).	Text
is sub-sampling?	Records if the sample is a sub-sample (Options: Yes, No). In many studies it was clearly stated that each sample was divided into two sub-samples, one was analysed and the other one was used as retained sample. In such cases the option "Yes" was selected. In some studies more than one honeybee hives (=replicates) were placed in each tunnel/glasshouse. At each sampling time samples were taken from each hive and these samples have been considered as sub-samples although it is not correct with the strict sense of the term "sub-sample".	List
Sub-sampling ID	The sampling ID is an identification code of each sub-sample and it was recorded if provided by the study.	List
Substance ID	Records the substance (active substance or metabolite) analysed in the sample. This column is a dropdown list that contains the identification codes of the substances recorded in "*STUDY*" sheet.	List
Residue levels of measured/analysed substance	Records the residue levels of the substance analysed in the sample.	Number
Residue levels of measured/analysed substance (unit)	It contains the Unit in which the residue levels of measured/analysed substance were expressed.	List
Type of residue levels measure (e.g. minimum, maximum, etc.)	Records the type of residue levels measured (Options: minimum, maximum, individual residue values per sample/subsample, etc.).	List
LOQ	Limit of quantification. The LOQ is specific for each study and analysed matrix.	Number
LOQ Unit	The unit in which the limit of quantification was expressed.	
LOD	Limit of detection. The LOD is specific for each study and analysed matrix.	Number
LOD Unit	The unit in which the limit of detection was expressed.	
Sampling After Application	Records whether the sample was taken before or after application (Options: Yes, No).	List

Columns	Description	Type
	The recording in this cell is always linked to the application ID mentioned in the next column (Application ID). For example, for studies with more than one applications, if sampling was carried out after the 1st and before the 2nd application, the option "No" has been selected in this cell and the Application ID of the second application was chosen in "Application ID" column. Alternatively, the option "Yes" was selected in this cell and the Application ID of the 1st application was chosen in "Application ID" column.	
Application ID	Records after which application the sample was taken. The field application ID takes values when a study ID is selected. No specific application was selected for the metabolites, since, as discussed and decided during the kick-off meeting of the project, no RUD calculations should be carried out for the metabolites.	List
Sampling After seed treatment	Records whether the sample was taken before or after sowing (Options: Yes, No). The recording in this cell is always linked to the seed treatment ID mentioned in the next column (Seed Treatment ID).	List
Seed Treatment ID	Records after which seed treatment the sample was taken. The field seed treatment ID takes values when a study ID is selected. No specific seed treatment was selected for the metabolites, since, as discussed and decided during the kick-off meeting of the project, no RUD calculations should be carried out for the metabolites.	List
Time between the pesticide application / sowing and sampling	Records the time between the pesticide (test item) application and the sampling time. The value "0" means that the sampling was conducted on the day of application. For samplings conducted on the day of application and after the application the option "Yes" was chosen in column "Sampling after application". For samplings conducted on the day of application and before the application the option "No" was chosen in column "Sampling after application".	Number
Time between the pesticide application / sowing and sampling (unit)	It contains the Unit in which the time between the application of the pesticide and the sampling event was expressed (Options: day, hour).	List
Sugar content of nectar	Records the quantity of sugar content of nectar in the sample.	Number
Sugar content of nectar (unit)	It contains the Unit in which the sugar content of nectar in the analysed sample was expressed.	List
Type of sugar content of nectar measure (e.g. minimum, maximum, etc.)	Scale of measurement of the sugar content of nectar (Options: minimum, maximum, etc.).	List
Time of sugar content sampling (e.g. morning, noon)	Time of nectar sampling (during the day) for the estimation of sugar content (Options: morning, noon, etc.).	List
Protein content	Records the content of protein detected in the sample.	Number

Columns	Description	Type
Protein content (unit)	It contains the unit in which the protein content was expressed.	List
Rainfall (l/m2/day)	Records the weather conditions (rainfall) during sampling. Rainfall measurement expressed in l/m2.	Number
Minimum relative air humidity (%)	Minimum relative air humidity during (or just before or after) the sampling (%)	Number
Maximum relative air humidity (%)	Maximum relative air humidity during (or just before or after) the sampling (%)	Number
Mean relative air humidity (%)	Mean relative air humidity during (or just before or after) the sampling (%)	Number
Minimum Temperature (° C)	Minimum temperature during (or just before or after) the sampling, expressed in ° C.	Number
Maximum Temperature (° C)	Maximum temperature during (or just before or after) the sampling, expressed in ° C.	Number
Mean Temperature (° C)	Mean temperature during (or just before or after) the sampling, expressed in ° C.	Number
Notes	Records any other remarks as regards the sampling and analytical determination of residue levels.	Text
RUDs (based on application rate)	The RUD values were calculated automatically as a quotient of the concentration (residue level in mg\kg) of the actual analysed samples (as captured in column M of the sheet "SAMPLING") divided by the relevant application rate (in kg/ha) used in each specific trial / plot / tunnel as indicated in column P of the sheet "APPLICATIONS". This column is only relevant for applications included in the "*APPLICATION*" Sheet and includes the RUD values derived by studies with spray, granule, drench application or drip irrigation. RUD values were calculated in all cases where the measured residue levels were above LOQ/LOD values. It is acknowledged that only some of these values can be further used, e.g. the worst-case values (derived from the highest measured residue values) for each trial/study/crop/matrix, values derived from samples taken shortly after the application, values derived from studies with a single application, etc. For further details please refer to Section 3.5.1 below. No RUD calculations were carried out for the metabolites.	Number
RUDs from seed treatment (based on seed dressing rate)	The RUD values were calculated automatically as a quotient of the concentration (residue level in mg\kg) of the actual analysed samples (as captured in column M of the sheet "SAMPLING") divided by the relevant seed loading / dressing rate (in mg a.s./seed) used in each specific trial / plot / tunnel as indicated in column Q of the sheet "*SEED_TREATMENTS*". This column is only relevant for treatments included in the "*SEED_TREATMENTS*" Sheet and includes the RUD values derived by studies with seed treatment when data for the seed loading / dressing rate were available. No RUD calculations were carried out for the metabolites.	Number
RUDs from seed treatment (based on	The RUD values were calculated automatically as a quotient of the concentration (residue level in mg\kg) of the actual analysed samples	Number

Columns	Description	Type
application rate)	(as captured in column M of the sheet "SAMPLING") divided by the relevant application rate (in kg a.s./ha) used in each specific trial / plot / tunnel as indicated in column P of the sheet "*SEED_TREATMENTS*". This column is only relevant for treatments included in the "*SEED_TREATMENTS*" Sheet and includes the RUD values derived by studies with seed treatment when data for the application rate were available. No RUD calculations were carried out for the metabolites.	

Step 7: "*CALCULATED_DT$_{50}$_DT$_{90}$*" worksheet population

The degradation kinetics assessment was performed following the FOCUS degradation kinetics, 2014 flowchart (Version 1.1), for persistence endpoints.

For certain studies and matrices where sufficient number of sampling time points and adequate residue values above LOQ/LOD are available, DT_{50} and DT_{90} for each matrix were calculated. Although for the vast majority of the relevant studies less than six sampling times were collected after the pesticide application, taking into consideration the complexity of the selected matrices and the absence of any guidance document for the test performance, $DT_{50/90}$ calculations were performed for all studies where a pattern of decline could be drawn. In several studies, only three sampling time points were considered, therefore, only SFO degradation kinetic fit was assessed for such cases.

In addition, tunnels/plots/trials that have were treated with approximately the same application rate in the same study, were considered as true independent replicates and the residue values per sample at each sampling interval were included individually for $DT_{50/90}$ calculations.

No DT50/DT90 calculations were performed for guttation fluid, since it is out of the objective of this specific project.

As regards the data handling, the recommendations as summarized in chapter 6.1 of FOCUS degradation kinetics (FOCUS, 2014, v1.1) were followed. In studies where no LOD was specified, the relevant samples below LOD and between LOD and LOQ were set to 0.5 x LOQ.

Kinetic modelling was carried out using CAKE v 3.1 (CAKE 2015). The raw data for the calculation of DT_{50} and DT_{90} values are presented in Appendix – H.

The output files of CAKE were captured in the sheet "*CALCULATED_DT$_{50}$_DT$_{90}$*", as follows:

Table 7: Data included in the relevant fields of the sheet "*CALCULATED_ DT$_{50}$_DT$_{90}$*".

Columns	Description	Type
Study ID	Identification code of the study, used in the sheet "*STUDIES*". The column Study ID is a dropdown list that contains the identification codes of the studies recorded in the "*STUDIES*" sheet.	List
Substance ID	Substance identification code, as indicated in "*SUBSTANCES*" sheet.	List

Substance name	Records the name of the active substance or the metabolite.	Text
Crop	Records the crop where the study was conducted.	
Trial ID	Identification code of the Trial as indicated in "*TREATMENT_GROUP*" sheet. In some cases different trials/plots/tunnels from each specific study were considered as true independent replicates, therefore, only one DT_{50} and DT_{90} calculation has been carried out for each matrix. In such cases the field "Trial ID" was left blank.	Text
Matrix	Type of sample taken: Nectar from bees, Pollen from bees, Nectar from flowers, Pollen from flowers, Honey from hive, etc.	List
Kinetic Model	Indicates the model that has been chosen for the calculation of DT_{50} and DT_{90} values.	Text
DT_{50} (days)	Calculated DT_{50} expressed in days.	Number
DT_{90} (days)	Calculated DT_{90} expressed in days.	Number
X^2	Records the Goodness of fit: Chi-squared error	Number
Visual assessment	Records the visual assessment of the fit (Options: excellent, very good, good, acceptable, non-acceptable).	List
k	Records the Rate constant of decline (1/days).	Number
k1 or alpha	Records the k1: Rate constant of decline of compartment 1 (1/days) (DFOP, HS) or a: shape parameter determined by coefficient of variation (FOMC).	Number
k2 or beta	Records the k2: Rate constant of decline of compartment 2 (1/days) (DFOP, HS) or β: location parameter (FOMC)	Number
g	Records the fraction of M0 applied to compartment 1 (DFOP, HS).	Number
Notes	Records any other useful remarks and comments.	Text
Number of sampling time points after application	Records the number of sampling time points after the application or sowing.	Number
Sampling time points after application	Records the sampling time points in days after application.	Text
Number of applications	Records the number of applications conducted in each specific trial.	Text
Reliability	Records the reliability of the calculated DT_{50} value, according to the criteria presented in Section 3.5.2.	Text
Justification	Records the justification for the allocation of the DT_{50} value in each reliability category.	Text

Step 8: "*FOLIAGE_RUD*" worksheet population

For the population of the "*FOLIAGE_RUD*" worksheet, data on residues on plant foliage were retrieved

from the "Residue data" section of finalized regulatory documents (i.e. DAR, RAR). Data presented in assessment reports that were under EU peer review the time the sheet was completed were not taken into account.

Only acceptable and reliable supervised residue studies used for MRL setting were considered. In cases where it was not possible to conclude on the acceptability of the studies and whether the respective data were used for MRL proposal (i.e. beta-cyfluthrin), all relevant studies were considered.

All relevant regulatory documents for all active substances under concern were screened. Exceptionally, for the active substance alpha-cypermethrin only the residue trials presented in Addendum to DAR (2003) were screened as no summary of the supervised residue trials reported in the DAR (1999) could be found.

Only residues in foliar parts of crops were considered, e.g. green material/whole plant/ear/straw for cereals, whole plant/remaining plant/plant (no cobs) for maize, whole plant/plant without pods or roots/green material for oilseed rape, head for cabbage, leaves for lettuce and kale, etc. Residues in fruits (e.g. of fruiting vegetables, orchards, grapes), tubers (e.g. of potato), curds (e.g. of cauliflower, broccoli), grain (e.g. of cereals), seeds (e.g. of oilseed rape, peas, beans), pods (e.g. for legumes, oilseed rape), buttons (Brussel sprouts) and root (carrot, beet) were not considered.

The residue data on plant foliage presented were derived from supervised residue trials performed in North, Central or Southern Europe in glasshouses and/or under field conditions. Where residue values for multiple PHI were reported, the highest value was selected.

All residue data presented were derived from foliar spray applications. For the active substance glyphosate, residue data on foliage were available from studies (accepted for MRL setting) where spraying performed before seedling planting. As residue levels measured in these studies were below LOD (<0.05 mg/kg), the respective data were not included in the database.

RUD (Residue per Unit Dose) values were calculated only in cases where foliar residues resulted from single applications. In studies where multiple applications were performed, the respective RUD values were considered of limited reliability due to the fact that the measured residue levels reflected both the total application rate and any degradation/dissipation processes occurred following each application.

Table 8 summarizes the data that were included in the sheet "FOLIAGE_RUD".

Table 8: Data included in the relevant fields of the sheet "FOLIAGE_RUD"

Columns	Description	Type
Substance Name	Name of measured/analysed substance.	Text
Substance CAS Number	Substance CAS number	List
Report/Study No	Identification code of the study, as indicated in the "Residues Section" in DAR/RAR	Text
Data Source (Regulatory Document)	Records the regulatory document (e.g. DAR, RAR) where the studies and relevant data were found.	Text
Location/Region of the Study	Records the exact location of the study.	Text

Columns	Description	Type
Country	Records the country where the study has been conducted.	List
Type of crop (commodity)	Records the type of crop used for each study (e.g. lettuce, cabbage)	List
Application rate (kg a.s./ha)	Records the rate of the pesticide a.s. used in each specific study.	Number
Growth stage at last treatment	Records the growth stage of the crop at the time of last application.	Text
Portion analysed	Records the portion of the crop that has been sampled and used for residue analysis.	Text
PHI (days)	Records the Pre-Harvest Interval, i.e. the time (in days) between the last pesticide application and the harvest of the treated crop (i.e. sampling).	Number
Residues (mg/kg)	Records the residue levels (in mg/kg) of the substance analysed in the sample.	Number
RUD	Records the RUD values, i.e. the amount of residues (mg) on 1 kg plant foliage normalized on an application rate of 1 kg a.s./ha.	Number
Notes	Records any other useful remarks and comments.	Text

Step 9: Creating the " *ALL_DATA_MERGED'* worksheet

Under this sheet the information recorded in the sheets '*STUDIES'*, '*TREATMENT_GROUPS*', '*APPLICATIONS'* , '*SEED_TREATMENT* 'and '*SAMPLINGS'* was combined. The fields contained under the above-mentioned sheets were sequentially brought together.

Through this worksheet, the user may review the main bulk of information stored in the database.

For future needs, the user has the possibility to repeat the procedure for the recreation of the worksheet following the steps described below. The latter is meaningful in cases when the database is updated with new information.

The data merging process is comprised of the following steps:

Step 1. Insert manually a blank worksheet named '*ALL_DATA_MERGED'*. If the worksheet already exists (from a previous version), the user should firstly delete the existing worksheet in question.

Step 2. Click on the worksheet named '*MERGED_BUTTONS*'. In this worksheet, the user should find six buttons named: (a) STEP 1-Merge SAMPLINGS, (b) STEP 2-Merge STUDIES, (c) STEP 3-Merge TREATMENT GROUPS, (d) STEP 4-Merge APPLICATIONS, (e) '*SEED_TREATMENTS'* and (f) STEP 5-Merge SUBSTANCES.

Step 3. Click on each of the abovementioned buttons sequentially. By clicking each button a procedure is initialised where the contents included under the worksheets 'SAMPLINGS', 'STUDIES', 'TREATMENT GROUPS', 'APPLICATIONS', 'SEED_TREATMENTS' and 'SUBSTANCES' are copied to the 'ALL_DATA_MERGED' worksheet. The user should not proceed with the execution of the subsequent steps if the process under the previous step has not been completed. Therefore, the user should check when executing each step of the process whether all information has been copied under the 'ALL_DATA_MERGED' worksheet.

2.5. Methodology of data analysis

The data analysis centres around four core objectives to investigate potential correlations between:

a) residue levels in pollen or nectar versus residue levels in plant foliage;

b) residue levels in pollen versus residue levels in nectar;

c) residue levels in pollen or nectar versus physical-chemical and environmental properties of the active substance (S, logPow, Koc);

d) residue decline in pollen or nectar versus residue decline in other environmental matrices.

The data analysis consists of a significant tool that could be employed in the risk assessment of pollinators under the framework of Regulation No 1107/2009 of the European Parliament and European Council considering the placing of plant protection products on the market.

To tackle the objectives, descriptive statistics, quantitative and inferential analysis were considered. Summaries and graphics of the variables of interest were primarily performed to give a visual understanding of the distribution of the variable values, and hypothesis tests and statistical analyses such as analysis of variance and correlation analysis, were performed to advocate the results. Detailed descriptions of the methods employed are given below.

To perceive the distribution of the residue in nectar, pollen and plant foliage, firstly a summary of the values was carried out that includes the minimum and maximum, first and third quartiles, the mean and median, and the standard deviation of the observations.

Graphically, boxplot were created. Boxplot is a diagram that provides an advantageous and convenient way of displaying the distribution of a dataset, based on the minimum, the first and third quartiles, the median and the maximum of the data. The first quartile marks the value below which the 25% of the observations are located in the ranked dataset. The third quartile respectively marks the value below which the 75% of the observations are located. In the same manner, the median is the value which divides the ranked dataset in two halves. The boxplot is basically composed of a rectangular box that spans from the first to the third quartile, thus indicating the interquartile range (IQR). The line inside the rectangle shows the median and the two vertical lines to the left and right sides of the box give the minimum and maximum values respectively that are not identified as outliers. Outliers are added in the boxplot as individual points, defined by employing Tukey's definition according to which an observation is an outlier if it falls 1.5*IQR above the third or below the first quartile.

The mean and median provide two considerably useful measures of central tendency, median being particularly advantageous for skewed data, and the standard deviation and interquartile range are employed as measures of dispersion.

Further, analysis of variance was performed to identify possible differences of the residue values between sampling matrices, type of crop and substances. Analysis of variance (ANOVA) is one of the most useful and most used statistical inference methods, as it provides a method of testing whether there exist statistical significant differences among group means. In a hypothesis test, a result is said to be statistically significant if it is unlikely to have occurred by chance, considering that the null hypothesis is true. The null hypothesis is rejected when the probability value (p-value) of the test is less than the significance level of the test, usually 5%. The p-value is the probability of observing the same or more extreme results as the observed ones resulting from the data available, when the null hypothesis is true. The null hypothesis in an ANOVA test is that all the groups are essentially random samples from the same population. Rejecting the null hypothesis implies then that the samples come from different populations.

In addition and complimentary to ANOVA, linear regression was performed, to assess the relationship between two variables and see which independent variable (regressors – explanatory variables) is

related to the dependent variable (response), for example nectar RUDs as a response and pollen RUDs as an explanatory variable, allowing to make predictions based on the regression line.

Correlation analysis was employed to evaluate the degree of the relationship between two variables. Furthermore, correlation analysis was performed between the residue values in nectar, pollen and plant foliage, residue in nectar and pollen with physical-chemical properties of the active substances, and between the residue decline values in pollen, nectar and plant foliage. If correlation between the two variables is detected, this means that when the one variable changes the other changes as well. This does not mean that change in one variable causes the change in the other variable, it means however that the two variables change in parallel, possibly due to the effect of another factor. In general, the correlation coefficient takes values between -1 and 1. A number close to these limits indicates strong negative or positive correlation respectively, whereas a correlation coefficient of 0 indicates that there does not exist a linear relationship between the two variables. The correlation analysis was conducted by employing the Pearson and Spearman correlation coefficients. On the one hand, the Pearson correlation coefficient evaluates the linear relationship between the two variables, that is a change in one variable is associated with a proportional change in the other variable. On the other hand, the Spearman correlation coefficient evaluates the monotonic relationship between the two variables, in which case the variables change together, but not necessarily at a proportional rate.

To test the hypothesis that the sets of the RUD values are different from each other subject to various characteristics (sampling matrix, type of crop etc.), we employed the well known two-sample t-test of the null hypothesis that the means of two populations are equal. In principal, the Student's t-test is employed once equality of population variances is assumed, while the Welch's t-test is employed if the equality of variances assumption fails. The Welch's unequal variances t-test is a modification of the Student's t-test that is more robust under the non equality of population variances and also under unequal sample sizes, while further it provides a more robust test against an analysis of variance (ANOVA). Although not mandatory, an equality of variances test was first conducted, testing the null hypothesis of homoscedasticity. In addition, we employed the Mann-Whitney test which is the non-parametric alternative to the t-test and does not depend on the underlying distribution, making it more robust and widely applicable.

3. Results

3.1. Screening of regulatory documents for the identification of relevant studies

The screening of the regulatory documents was carried out as described under point 2.1 for the following active substances:

- 29 AIR II substances listed in Reg (EC) No 1141/2010
- 150 AIR III substances listed in Reg (EC) No 686/2013
- 205 New Active Substances (NAS) listed in the EU Pesticide Database

Finally, 314 active substances were screened, since sixteen (16) and fifty four (54) of the New Active substances are also listed in Reg (EC) No 1141/2010 (AIR II) and Reg (EC) No 686/2013 (AIR III) respectively.

The detailed list of the active substances as well as the screened regulatory documents is presented in Appendix B.

In addition, unpublished studies were submitted by chemical industry to EFSA and provided to the contractor. These studies were conducted by ADAMA AGRICULTURAL SOLUTIONS, BASF, Bayer CropScience, DuPont Crop Protection and Sumitomo Chemical Agro.

Therefore, thirty (30) substances were identified with relevant studies conducted after 2010 and these substances are presented in Table 9 below.

Table 9: List of active substances with relevant studies

	Active substance	Category of substance (AIR II / AIR III / NAS)	Number of studies from regulatory documents	Number of studies submitted to EFSA
1	2,4 D	AIR II	1 study (RAR 2014)	
2	Acetamiprid	AIR III	1 study (RAR 2015)	4 studies (ADAMA)
3	Alpha-cypermethrin	AIR III		2 studies (BASF)
4	Beta- cyfluthrin	AIR III		2 studies (ADAMA)
5	Boscalid	AIR III		2 studies (BASF) (formulation with dimoxystrobin + boscalid)
6	Chlorantraniliprole	NAS		3 studies (DuPont)
7	Clothianidin	AIR III	15 studies (DAR addendum confirmatory info BCS) 8 studies (DAR addendum confirmatory info Sumitomo)	4 studies (Sumitomo) (3 of these studies are presented also in DAR addendum confirmatory info Sumitomo)
8	Cyantraniliprole	NAS	29 studies (DAR 2014)	
9	Cyclaniliprole	NAS	6 studies (DAR 2015)	
10	Dimoxystrobin	AIR III		Common studies (2) (BASF) with boscalid
11	Emamectin	NAS	2 studies (Registration report REVIVE)	
12	Ethephon	AIR III		1 study (Bayer)
13	Fluopyram	NAS		2 studies (Bayer) (formulation with fluopyram &

	Active substance	Category of substance (AIR II / AIR III / NAS)	Number of studies from regulatory documents	Number of studies submitted to EFSA
				trifloxystrobin)
14	Flupyradifurone	NAS	13 studies (DAR 2014)	
15	Folpet	AIR III		1 study (ADAMA)
16	Gamma-cyhalothrin	NAS	2 studies (DAR Addendum 2013) 2 studies (Registration Report: Nexide CS)	
17	Glyphosate	AIR II	1 study (RAR 2013)	
18	Indoxacarb	AIR III		7 studies (DuPont)
19	Iprodione	AIR III	2 studies (RAR 2015)	
20	Metconazole	AIR III		1 study (BASF) (formulation with pyraclostrobin & metconazole)
21	Oxamyl	AIR III	1 study (Registration Report: Oxamyl 10% SL)	
22	Pymetrozine	AIR II	9 studies (RAR 2013)	
23	Pyraclostrobin	AIR III		Common study (BASF) with metconazole)
24	Spirotetramat	NAS	1 study (DAR addendum 2013) 1 study (Registration Report: MOVENTO 150 OD)	
25	Tau-fluvalinate	AIR IV		1 study (ADAMA)
26	Tebuconazole	Existing substance		1 study (Bayer)

	Active substance	Category of substance (AIR II / AIR III / NAS)	Number of studies from regulatory documents	Number of studies submitted to EFSA
27	Thiacloprid	AIR III		1 study (Bayer)
28	Thiamethoxam	AIR III	1 study (Confirmatory data 2016)	
29	Thiram	AIR III	1 study (RAR 2015)	
30	Trifloxystrobin	AIR III		Common studies (2) (Bayer) with fluopyram

3.2. List of relevant studies

As described under point 3.1, thirty (30) active substances (a.s.) were found with relevant studies. For some of them (e.g. 2,4 –D, folpet, glyphosate, Metconazole, etc) only one study was identified. On the contrary, for other substances numerous studies were found (i.e. Cyantraniliprole, Clothianidin, Flupyradifurone, Pymetrozine and Cyclaniliprole).

Most of the studies (96 studies) were identified from the screened regulatory documents mainly from DAR, RARs and confirmatory data. Since only the summaries of these studies were available in the regulatory documents, the original study reports were collected and used for the evaluation of each study and the population of the database. These study reports were submitted to the Greek Ministry of Rural Development & Food General Directorate of Sustainable Plant Produce, Directorate of Plant Produce Protection and they were provided to the contractor in order to be used for this project.

In addition (as previously mentioned), thirty two (32) unpublished studies were submitted by chemical industry to EFSA and provided to the contractor. These studies were conducted by ADAMA AGRICULTURAL SOLUTIONS (8 studies), BASF (5 studies), Bayer CropScience (5 studies), DuPont Crop Protection (10 studies) and Sumitomo Chemical Agro (4 studies). A partial overlapping has been observed since 3 studies submitted by Sumitomo to EFSA were also found in regulatory documents.

Therefore, in total, one hundred and twenty five (125) studies were collected for the aforementioned thirty active substances. It is noted that only three (3) studies were found with estimation of sugar content of nectar and protein content of pollen for the active substances alpha-cypermethrin, boscalid and iprodione. All identified studies are presented in a detailed Table in Appendix – C. Furthermore, the identified studies are listed in an ENDNOTE library file and be attached in Appendix – D.

As regards the screening of international (ouside Europe) organisations and evaluating authorities' websites some studies were identified. More specifically in "White Paper in Support of the Proposed Risk Assessment Process for Bees" submitted to the FIFRA Scientific Advisory Panel for Review and Comment in 2012, twenty (20) study summaries from unpublished, registrant- submitted studies and ten (10) study summaries from open scientific literature were found. Some of these studies have been already considered and included in the Appendix F of EFSA bees GD.

Furthermore, residues data of sulfoxaflor in hive materials from field studies conducted in cotton, pumpkin and *Phacelia* are presented in EPA regulatory documents (2016 Addendum to the Environmental Fate and Ecological Risk Assessment for Sulfoxaflor Registration). However, since only short study summaries and not the detailed study reports were available, these studies cannot be evaluated and included in the developed database. Nevertheless, since the number of the identified studies from other sources was much more than the expected, it was considered than none of these studies should be included in the database and no further search was performed.

3.3. Selection of the studies to be included in the database

As described above, one hundred and twenty-five (125) studies were identified as relevant for this project. The number of identified studies was much higher than the initially expected one, since according to the Technical Offer and the respective Service Contract approximately forty-five (45) studies were expected to be found. Therefore, during the 1st stage of the project (June 2016 – January 2017) a prioritization procedure was carried out for the selection of the most appropriate studies to be included in the database. The studies that fulfilled one or more of the following criteria were finally selected:

- Studies with spray application were preferred over seed treatment or granule application
- Studies with adequate number of sampling time points to get valuable information for residue decline and/or maximum residue values
- Studies with sampling directly from plants or from bees
- Studies with many analysed matrices
- Studies with residue calculations for pollen & nectar and for foliage (in the same study if possible)
- Studies with estimation of protein and sugar content in pollen and nectar respectively

The final decision for the studies to be initially included in the database was taken in collaboration with EFSA. The selected studies that were included in the database during the 1st stage of the project implementation were marked in khaki in the list of identified and selected studies presented in Appendix-C.

Following the amendment No1 of the Service Contract the 80 additional studies that have been retrieved were also included in the database.

3.4. Evaluation of the selected studies

Each identified study was assessed according to the assessment protocol that has been developed based on the principles of Appendices G and S of the EFSA GD on bees (EFSA, 2013). It is acknowledged that due to time constrains an in-depth evaluation of the studies was not possible. However, the elements and parameters that were considered to be the most important for the quality of each study have been assessed. Furthermore, the evaluation was focused on parameters / elements relevant for this specific project; i.e. initial/maximum residue levels in pollen and nectar sampled in front of the hives (from bees captured in front of the hives/pollen traps fixed to the hive) or sampled directly from the crop and calculated DT_{50}/DT_{90} values for the above mentioned matrices.

Therefore, the following criteria were applied only to samples of pollen and nectar collected in front of the hives (from bees) or directly from the crop. The data collected for other matrices were used as supplementary information only and they have not been considered for the categorization / characterization of the studies.

Based on the outcome of the evaluation, the studies were allocated in four (4) categories as follows:

Category I: Fully fits to the purpose of this project

Not important deviations from EFSA GD recommendations were observed in the studies included in this category. The maximum residues derived by these studies were captured with very high certainty and clear decline were observed. Additionally, these studies were conducted in a typical agricultural site for the EU with no extreme weather conditions (e.g. heavy rain, temperatures, etc).

Category II: Fits to the purpose of this project with minor notes

This category contains studies with deviation from EFSA GD recommendations such as:

- Fewer than recommended sampling time points.

  According to the assessment protocol, at least 5 samplings should be conducted: one day before application, immediately after application and on at least 3 additional sampling times. The performance of only two sampling events was considered as minor deviation; the first sampling should had occurred immediately after application, i.e. 0DAA or 1DAA.

- In case of pollen and nectar collection from plants, the number of sampling locations was lower and/or the distribution of sampling locations in the treated field deviated from the recommendations given in the assessment protocol. This deviation can be considered as minor provided that the area(s) of the treated field sampled is (are) representative of the entire field.
- No adequate data for the test organisms (e.g. health status of bees, strength of colonies) were available.
- Number of samples (e.g. bees or plants/flowers) was lower than the recommended by the assessment protocol. This deviation was considered as minor provided that the amount of the collected sample was sufficient to meet the requirements of analytical procedures.
- Insufficient reporting of the weather or other atypical agricultural site conditions that may lead to uncertainty of the collected data.

The above mentioned deviations were considered to be "minor deviations" for the studies used for the purpose of this project.

Category III: Fits to the purpose of this project with some shortcomings

This category contains studies with deviation from EFSA GD recommendations such as:

- Only one sampling after application was carried out, even if this sampling was conducted immediately after application, i.e. 0DAA or 1DAA (unknown uncertainty on whether the maximum was captured). In such case the study was considered to be "reliable with major deviations".
- Very poor reporting of the weather or other atypical agricultural site conditions that may lead to uncertainty of the collected data.

Category IV: Not fit to the purpose of this particular project

This category contains studies with deviation from EFSA GD recommendations such as:

- Very limited information regarding the field phase of the trials (including the growth stage of the crop at the time of application, weather conditions, etc) was provided; therefore, possible deviations from EFSA GD was not possible to be identified.
- The first sampling in the study was carried out later than the 1st day after application. In this case, the initial RUD values could not be considered to be the worst case, therefore they could not be further used establishing initial/maximum residue level. The derived RUD values were reported only as supplemental information. Furthermore, DT_{50} values derived from these

studies were also reported only as supplemental information as in lack of measurement from the initial part of the decline brings considerable uncertainties on the possible shape of the decline curve.

The RUD and DT_{50} values derived by category I, II and III studies were further used for the purpose of this project e.g. for data analysis. On the contrary, the RUD and DT_{50} values derived from studies of category IV were not further used for data analysis. Additionally, RUD and DT_{50} values from studies characterized as "Not fit to the purpose of this particular project (?)" have not been considered for data analysis. However, these studies maybe contain information that could be used for the revision of the EFSA bee guidance document; therefore EFSA will take the final decision whether some data from these studies could be considered as reliable.

During the evaluation procedure the evaluation check list was filled in for each study and all completed evaluation check lists are presented in Appendix E.

3.5. Completed database

All relevant data from each one of the identified studies have been extracted and captured in the database as described in Section 2.4 of the present Report. It is noted that the study reports from four studies (study IDs: TK 0021089-FS-11, TK 0021089-11, 13 10 48 021 B, 13 10 48 084 B) was not possible to be found, therefore these studies were not finally included in the database. The completed database is presented in Appendix F.

A brief description summarizing the main points of each study that have been included in the database is presented in Appendix G.

According to the contract "*residue levels, expressed in mass/mass (i.e. mg/kg) measured in relevant matrices for each time point in relevant and unified statistical units, such as minimum, maximum, average, median, 90th %-tile and standard deviation. Detailed data for each time point for sample/subsample will be reported as well in a separate database (supplementary database).*" Since the detailed data already have been included in the database, the minimum, maximum, average, *etc* values were calculated from respective residue levels and were included in a separate spreadsheet entitled "Residues data & statistical units". These calculations were conducted within each reliable study, matrix and trial only for the samples that can be considered as "true replicates" (e.g. samples taken on the same time points after the application). This file contains most of the data presented in "*SAMPLING*" spreadsheet of the main Database and also the calculated statistical values in columns AA to AF. The sampling ID presented in column A can be used as a unique identification code for each sample in order to retrieve any other important and useful information from the main Database which is not included in the "Residues data & statistical units" file.

3.5.1. RUD values

RUD values were calculated for each analysed sample and included in the database (worksheet "SAMPLINGS" columns AN to AP).

For spray applications RUDs values were calculated as a quotient of the concentration (residue level in mg\kg) of the actual analysed samples divided by the relevant application rate (in kg/ha) used in each specific trial / plot / tunnel. No RUD calculations were carried out for the metabolites.

For seed treatment, the RUD values were calculated based on seed dressing rate or/and application rate. The RUD values based on seed dressing rate were calculated as a quotient of the concentration (residue level in mg\kg) of the actual analysed samples divided by the relevant seed loading / dressing rate (in mg a.s./seed) used in each specific trial / plot / tunnel. In some cases the seed dressing/loading rate was not included in the study report but it was calculated considering the application rate (kg a.s./ha) and the seeding rate (seeds / ha). The RUD values based n application rate were calculated as a quotient of the concentration (residue level in mg\kg) of the actual analysed samples divided by the relevant application rate (in kg a.s./ha) used in each specific trial / plot / tunnel.

It is acknowledged that although RUDs were calculated for each collected sample included in the database, only some of these values were considered reliable and appropriate in order to be further used for (i) data analysis performed in the frame of this project and (ii) enrichment of the data set of RUD values already available in Appendix F of the EFSA bees GD [EFSA Journal 2013:11(7): 3295] and used for pollinators risk assessment. A detailed discussion for the final selection of reliable RUD values was conducted during the final meeting of the project and the following rules for the selection of the most appropriate values were agreed:

- RUDs values from different study, different matrix and different trials were kept separately.
- RUDs values for the same matrix (e.g. pollen) collected with different mode of sampling (e.g. pollen from the plant, pollen from the bee or pollen from combs) were kept separately.
- RUDs were calculated from residue level ≤ LOQ assuming that the measured residue levels are equal to LOQ (as a worst-case assumption).
- RUDs were calculated from residue level ≤ LOD assuming that the measured residue levels are equal to LOD (as a worst-case assumption).
- RUD values were calculated only for studies/matrices with 1st sampling point shortly after the application (on 0DAA or on 1DAA).
- RUD values were calculated even for studies with more than one application.
- When several measurements of residues for the same matrix were available within a trial (e.g. from different sampling time points), only the highest RUD value was selected and considered for the data analysis.

Thereafter, the RUD values from the database were collected in a separate file entitled "Evaluated and selected RUDs" and they were evaluated taking into consideration the above agreed points. RUD values from studies with different application method were collected in separate worksheets i.e.

- RUDs from studies with spray application
- RUDs from studies with seed treatment
- RUDs from studies with granule application

RUDs from studies with other application methods (e.g. drip irrigation, drench and combination of seed treatment and spray application or granule and spray application) were not included in this file, since we consider that the calculation of these RUDs were out of the objectives of this specific project. However, since the RUDs values derived from all relevant studies have been calculated in the Database, they can be evaluated and used by EFSA if needed.

The "Evaluated and selected RUDs" file includes most of the data as presented in "*SAMPLING*" worksheet of the database (except of data regarding the sugar and protein content and the weather conditions) and in addition the information illustrated in the following Table.

Table 10: Data included in the relevant fields of "Selected RUDs" file

Columns	Description	Type
RUD notes	Records important notes as regards each RUD value e.g. value derived by residue value ≤ LOQ.	Text
Reliable RUD value?	Indicates whether the RUD value is considered reliable (YES/NO)	Text
Justification	Records the justification for keeping or not each RUD value in the list of reliable values	
RUD value used for data analysis	Indicates whether the RUD value have been considered for the data analysis. From all reliable RUD values only the worst case values were used for the data analysis. Furthermore, the only relevant matrices for data analysis were the following: - Pollen from traps - Pollen from bees - Pollen from flower - Nectar from bees - Nectar from flower Therefore, only reliable RUD values from these matrices were used for data analysis.	Text
Number of applications	Records the number of applications that have been performed in the trial/study from which the RUD value derived.	Text
Type of higher tier study	Records the type of higher tier study from which the RUD value derived (i.e. field study, tunnel, glasshouse)	Text
Direction of application	Records the direction of application that have been performed in the trial/study from which the RUD value derived.	Text

Therefore, the final list of calculated RUD values and the final decision as regards their reliability along with the respective justification is presented in the file "Evaluated and selected RUDs".

The most robust data set was derived by studies with spray foliar application of pesticides. Many reliable RUD values were calculated for nectar and pollen for several active substances.

For granule applications the only reliable RUD values were derived from studies with the active substance clothianidin. The RUDs in pollen collected from different sources (i.e. bees, pollen traps and flowers) were comparable and no decline was observed in samples taken in different time points after the start of flowering.

For seed treatment not reliable RUD values were derived for pollen or nectar, since the only available residue data were collected from not reliable studies i.e. studies with inadequate reporting of the study design in the study report or studies with only one sampling time point. However it is acknowledged that especially for seed treatment, the RUD values from studies with only one sampling time point during flowering could be used, since a residue decline is not expected to be observed during the flowering period of the crop. Nevertheless, the further use of these data can be decided by EFSA.

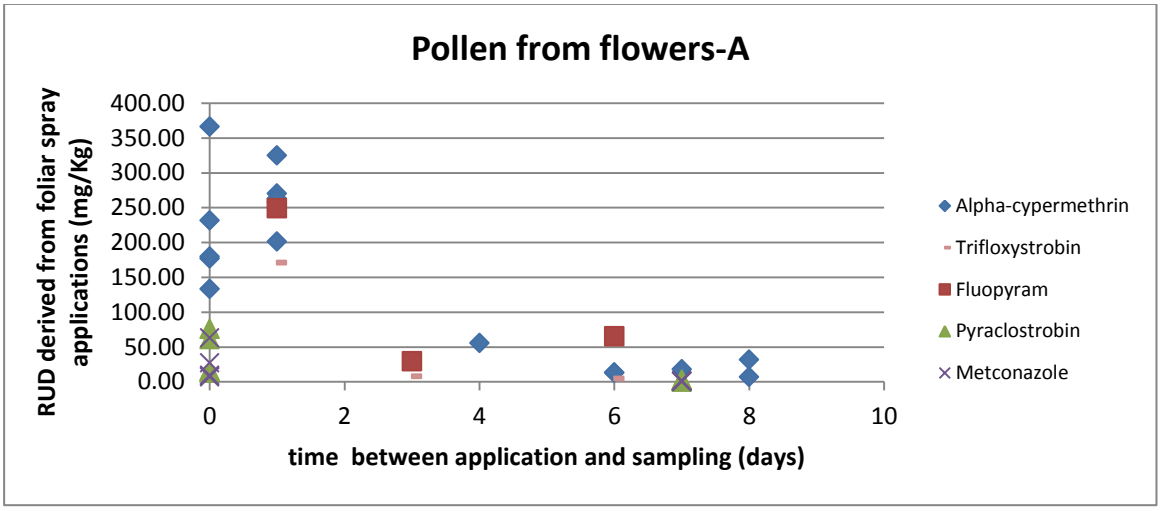
RUDs decline in each analysed matrix

The following graphs present the reliable RUD values from all available studies versus time (in days) per analysed matrix. Each active substance was presented separately in the graphs. However, a more detailed analysis of the collected data as regards the RUD values in different matrices is presented in Section 4 of this report (Data analysis results).

Spray applications

Pollen from flowers

For pollen collected from flowers, the highest RUD values were observed immediately after the application on 0DAA or 1DAA. Some active substances (e.g. alpha-cypermethrin, fluopyram) exhibit high RUDs (>200 mg/kg) on 0DAA and 1DAA, while the initial worst case RUDs for other substances (e.g. dimoxystrobin, boscalid, cyantraniliprole) were significantly lower (< 55 mg/Kg). In all cases a fast decline was observed and the RUD values calculated from samples taken later than the 2nd day after application were much lower than the initial values.



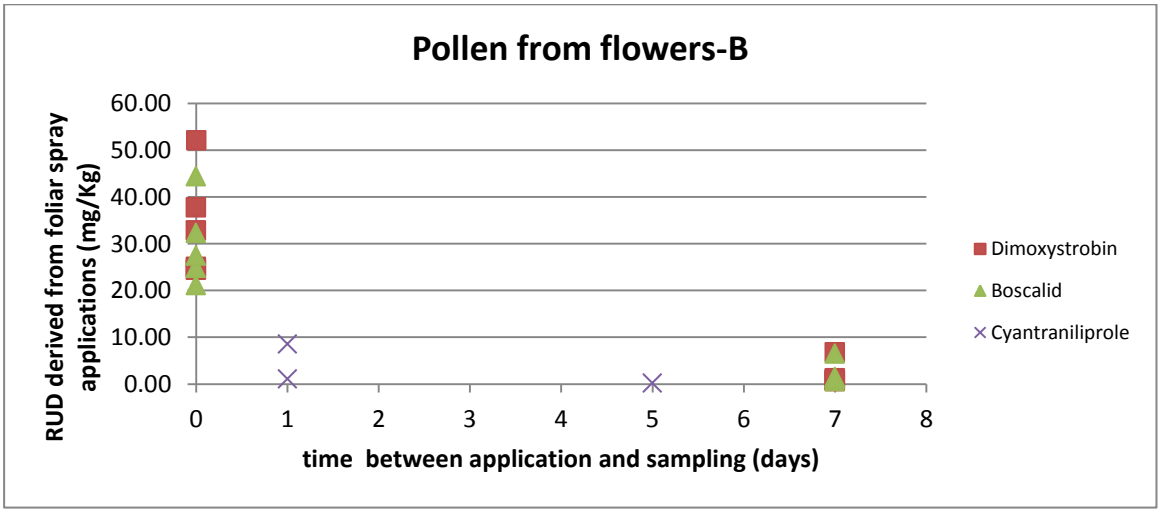
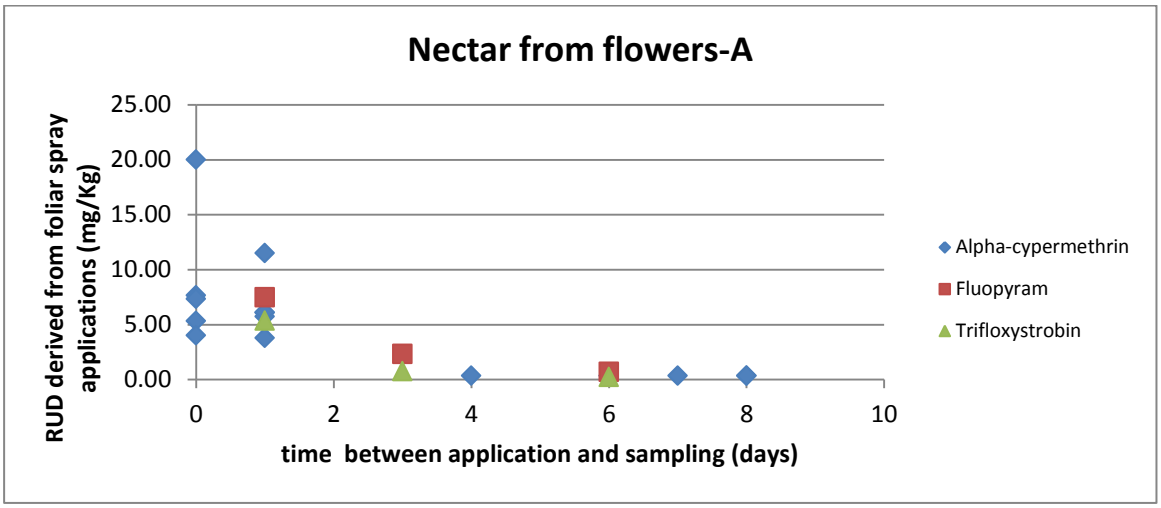


Figure 2: RUD values in pollen collected from flowers versus time between application and sampling.

Nectar from flowers

The highest RUD values in nectar collected from flowers were observed on 0DAA and 1DAA. Similarly with the pollen samples collected from flowers, some active substances (e.g. alpha-cypermethrin, fluopyram, trifloxystrobin) exhibit high RUDs (>5 mg/kg) in nectar samples on 0DAA and 1DAA, while the initial worst case RUDs for other substances (e.g. metconazole, boscalid, cyantraniliprole, pyraclostrobin) were significantly lower (< 2.5 mg/Kg). Low RUD values were observed in samples taken after the 2nd day after application. As it was expected, the RUDs in pollen collected from flowers, were much higher than the respective RUDs in nectar collected from flowers.



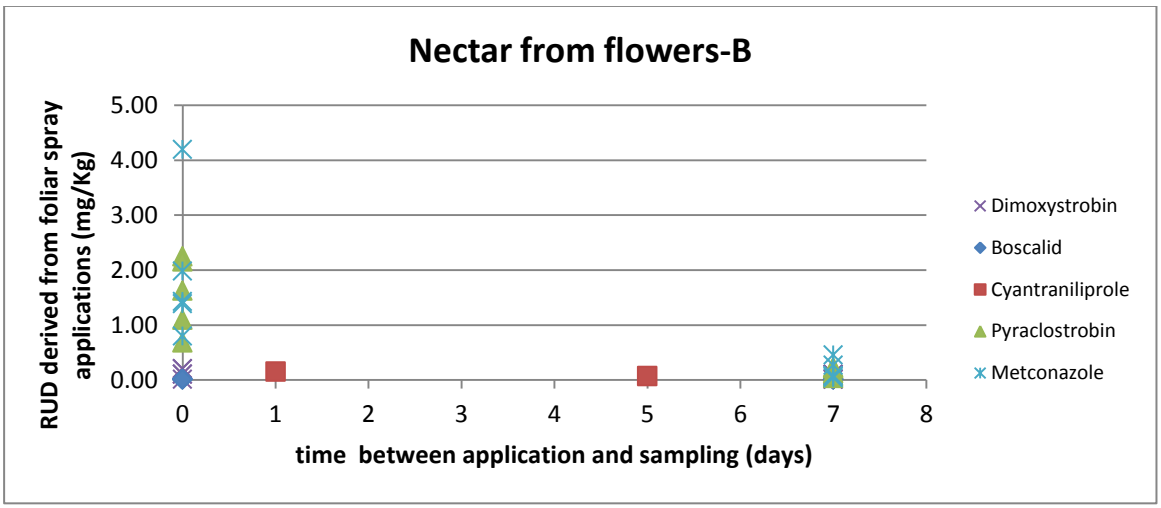
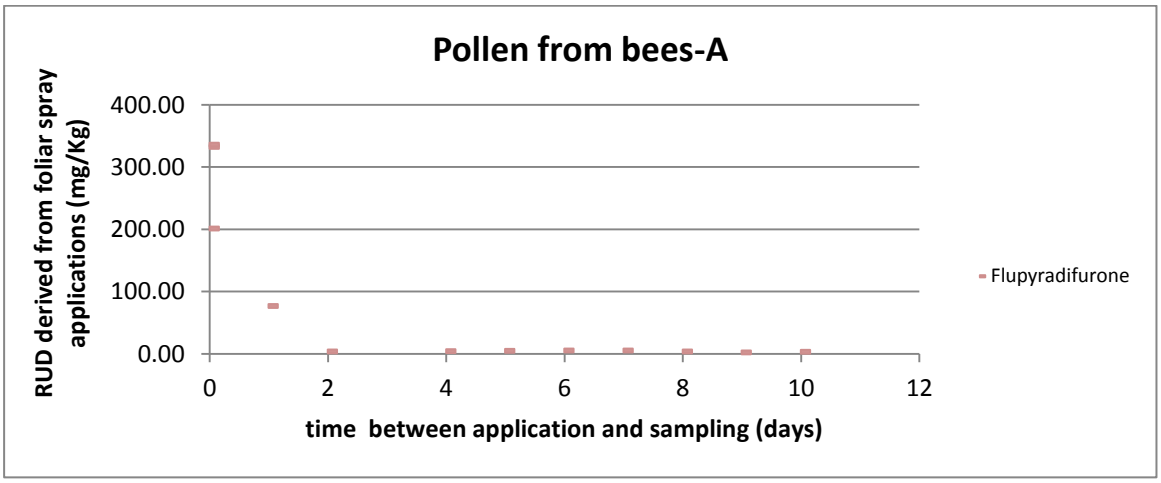


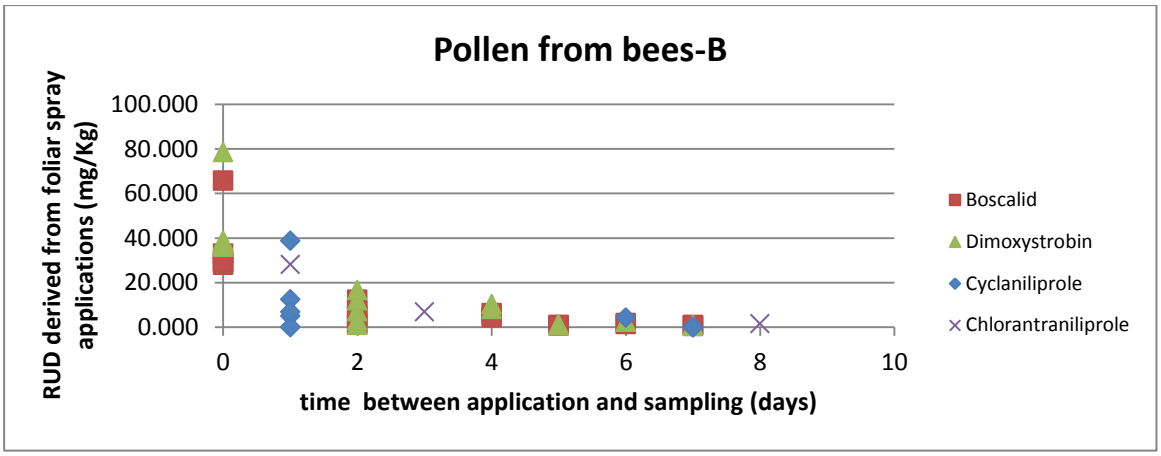
Figure 3: RUD values in nectar collected from flowers versus time between application and sampling.

Pollen from bees

The highest RUD values were observed immediately after the application on 0DAA or 1DAA. Three categories of active substances can be defined as regards the initial worst case RUD values. The

highest initial RUDs were observed in pollen collected from bees for the active substance flupyradifurone (> 200 mg/kg). Lower initial RUDs were calculated for boscalid, dimoxystrobin, cyantraniliprole, chlorantraniliprole (20-80 mg/kg approximately), while the lowest initial values (≤10mg/kg) were identified for beta-cyfluthrin and indoxacarb. In all cases significant decline in RUD values was observed in samples collected later than the 2DAA.





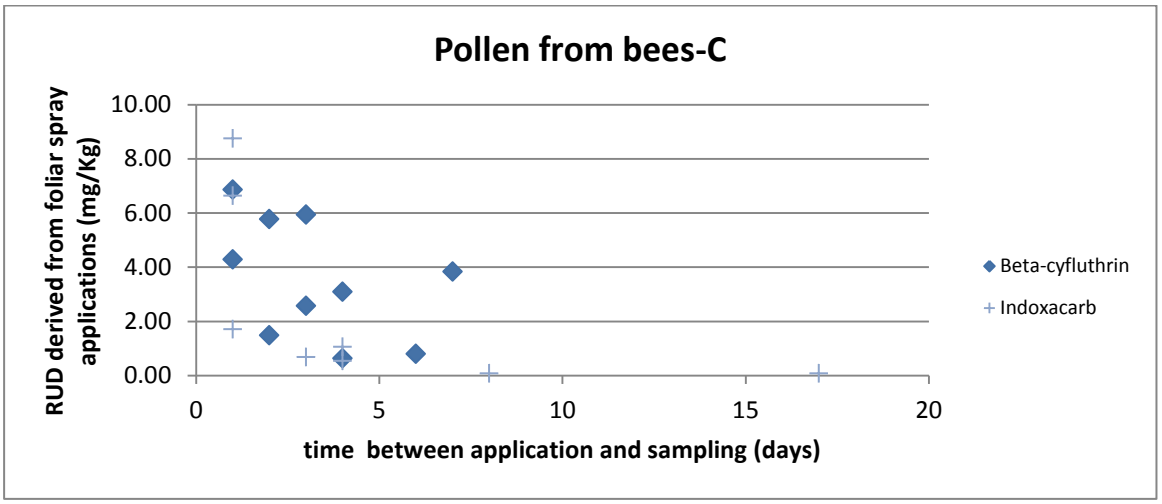
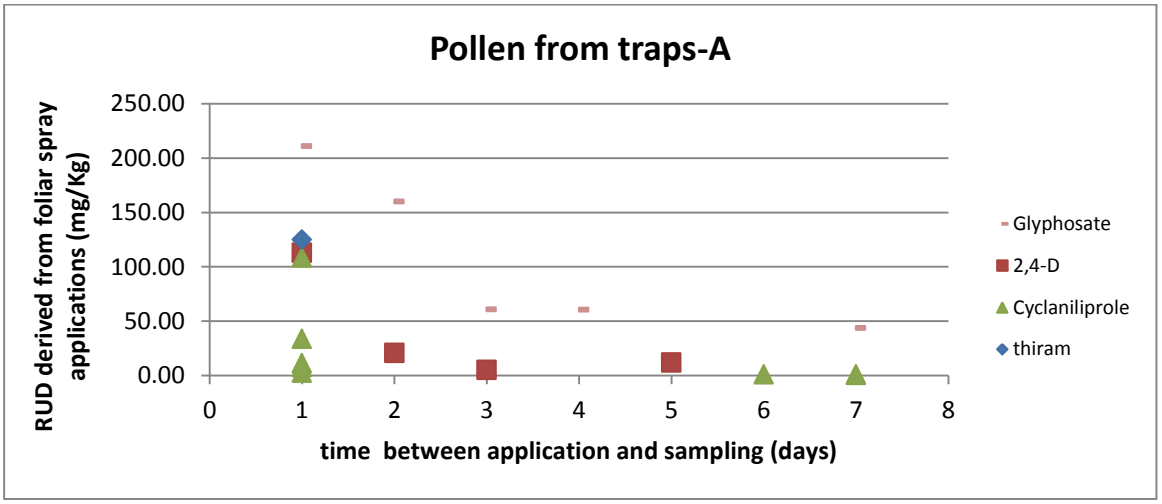


Figure 4: RUD values in pollen collected from bees versus time between application and sampling.

Pollen from traps

For pollen collected from traps, the highest RUD values were observed immediately after the application on 0DAA or 1DAA. The highest initial RUDs were observed in studies conducted with thiram, glyphosate, cyclaniliprole and 2,4 D (>100 mg/kg). Other active substances (i.e. alpha-cypermethrin, iprodione, cyantraniliprole, gamma-cyhalothrin and spirotetramat) exhibit lower initial RUD values (<50 mg/kg).



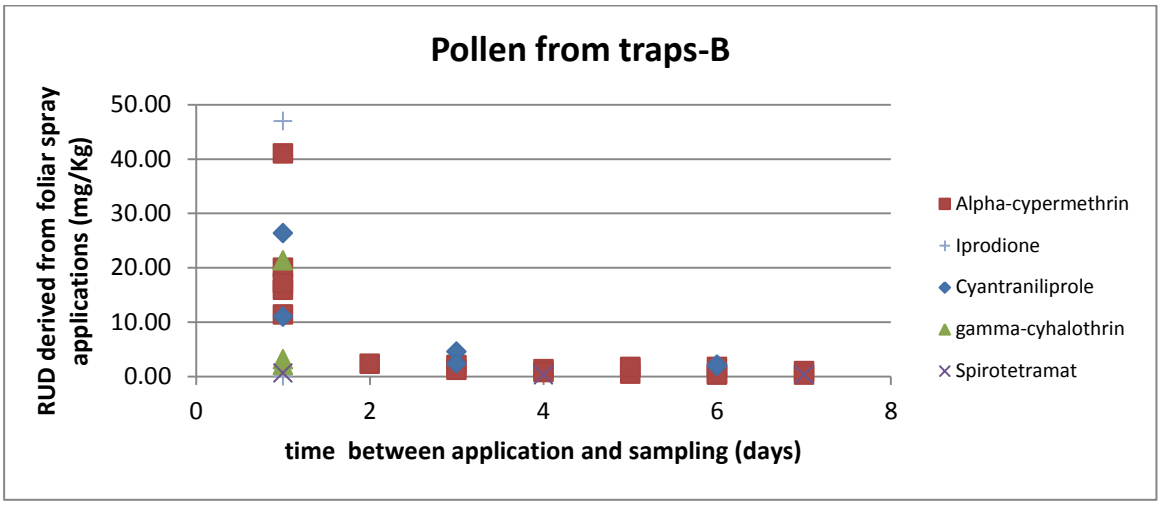
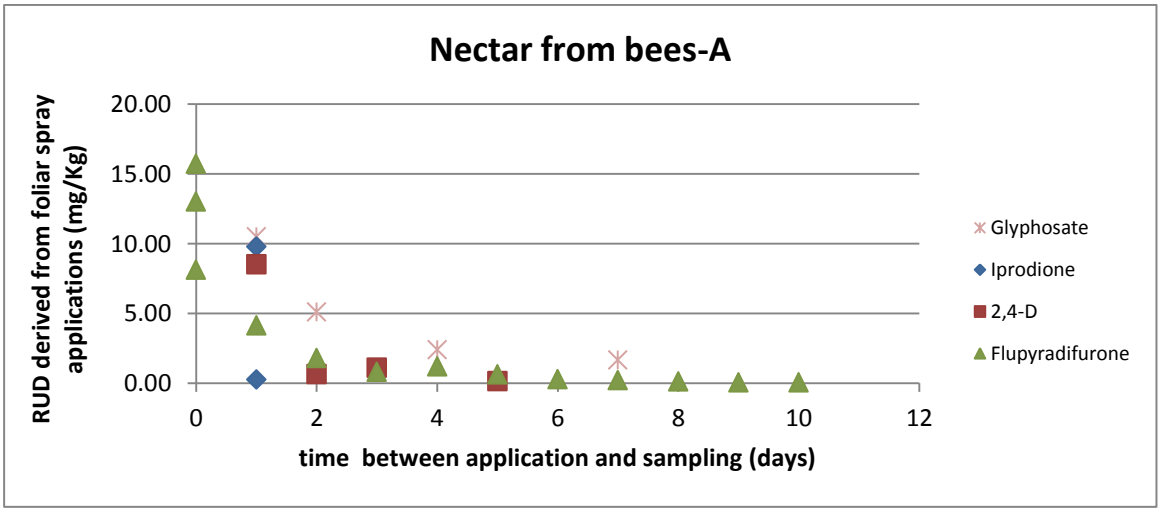


Figure 5: RUD values in pollen collected from traps versus time between application and sampling.

Nectar from bees

For nectar collected from bees, the highest RUD values were observed immediately after the application on 0DAA or 1DAA. Some active substances (i.e. glyphosate, 2,4-D, iprodione and flupyradifurone) exhibit initial RUDs values at approximately 10 mg/kg, while lower initial RUD values (≤ 5 mg/kg) were observed for other substances (i.e. Alpha-cypermethrin, beta-cyfluthrin, boscalid, dimoxystrobin, chlorantraniliprole, cyantraniliprole, gamma-cyhalothrin, cyclaniliprole, thiram). In all cases significant decline in RUD values was observed in samples collected later than the 2DAA.



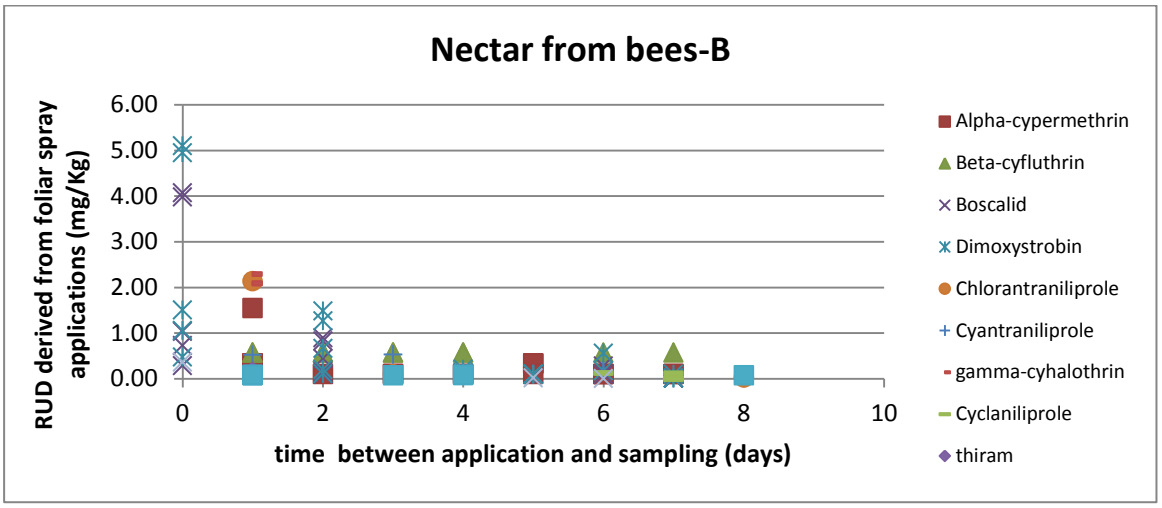


Figure 6: RUD values in nectar collected from bees versus time between application and sampling.

Granule applications

For granule applications the only reliable RUD values were derived from studies with the active substance clothianidin. As illustrated in figure 7 below, the RUDs in pollen collected from different sources (i.e. bees, pollen traps and flowers) were comparable and no decline was observed in samples taken in different time points after the start of flowering.

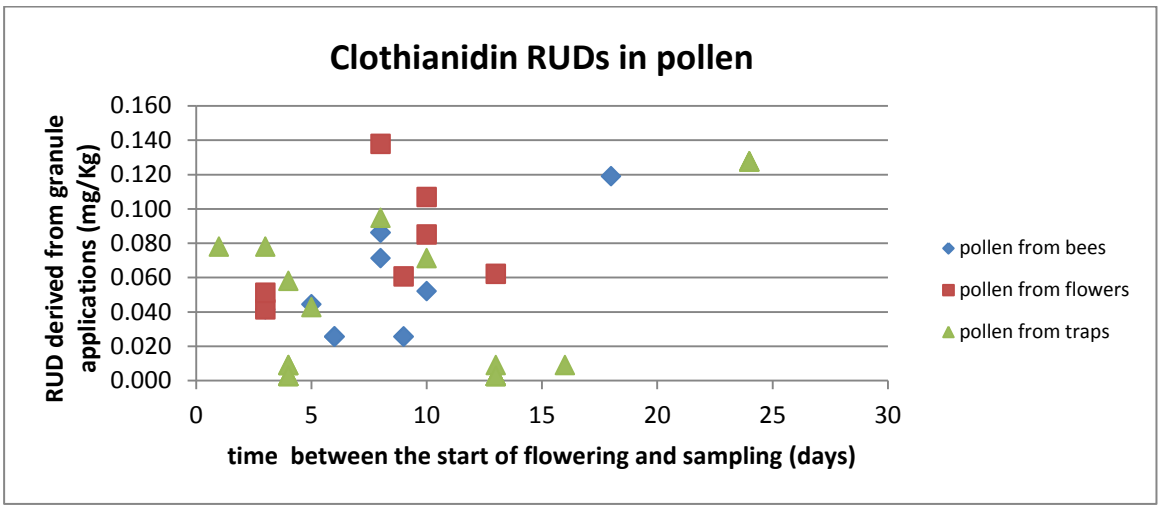
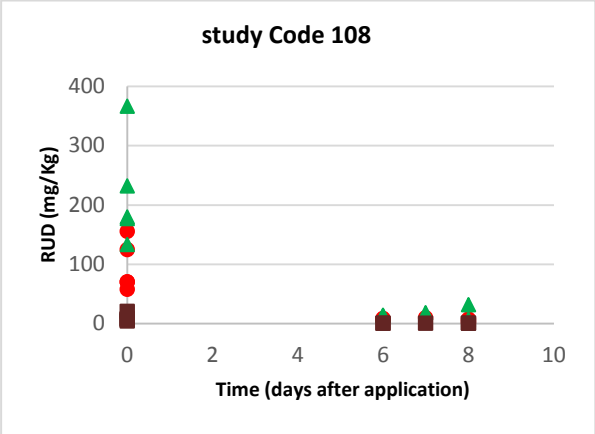


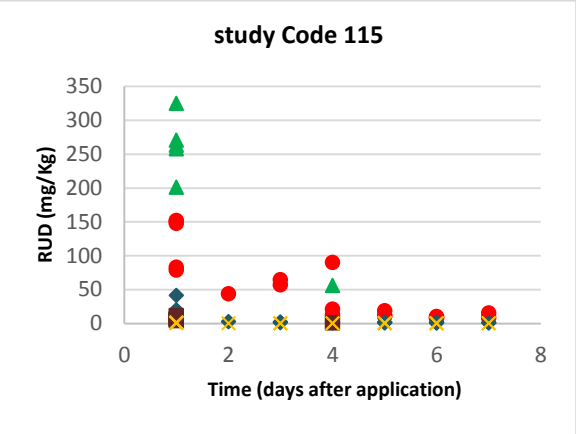
Figure 7: Clothianidin RUD values in pollen collected from bees, traps or flowers versus time between the start of flowering and sampling.

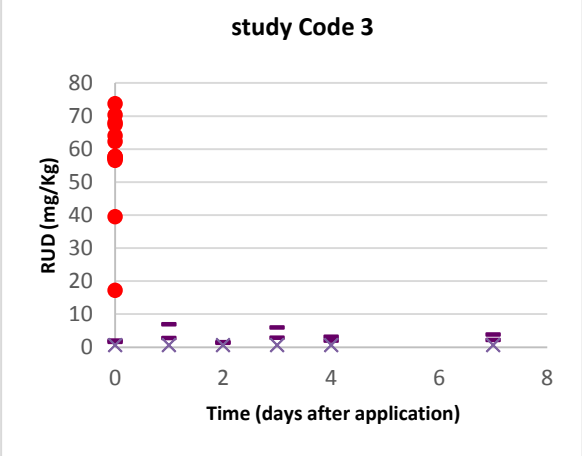
Comparison of RUDs in flowers and nectar or pollen

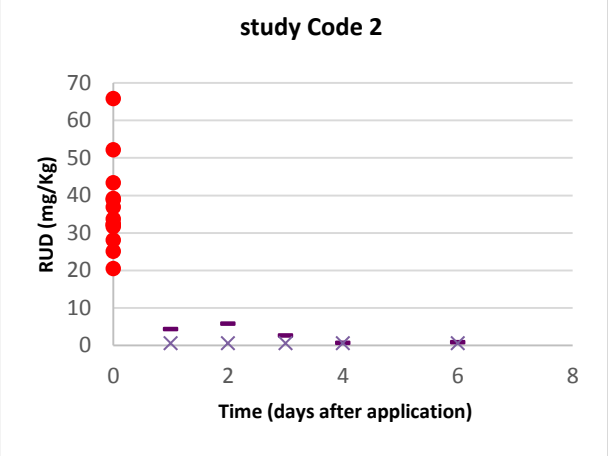
The following graphs present the reliable RUD values in flower, nectar (from bees or/and flowers) and pollen (from bees, traps and/or flowers) derived from each study with spray foliar application. These graphs where used in order to compare the RUDs in different matrices in each specific study.

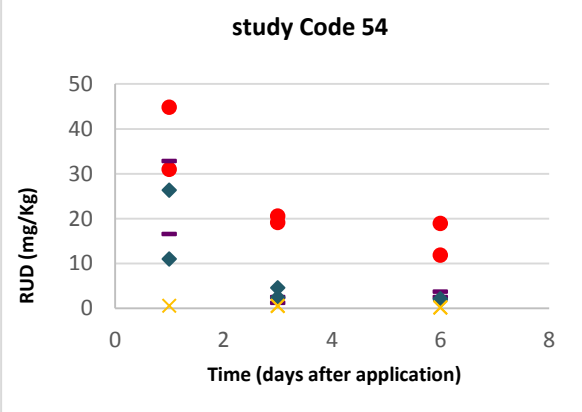
A more detailed analysis of the collected data is presented in Section 4 of this report (Data analysis results).

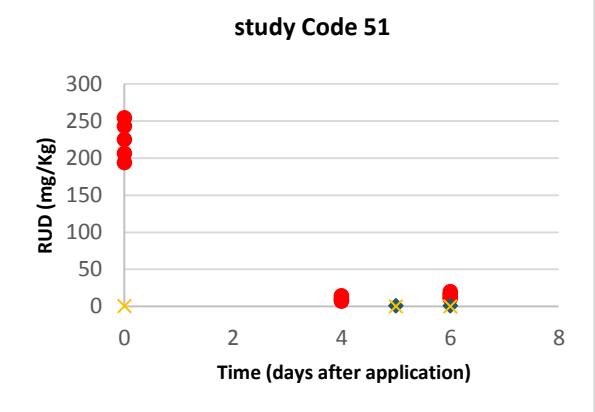


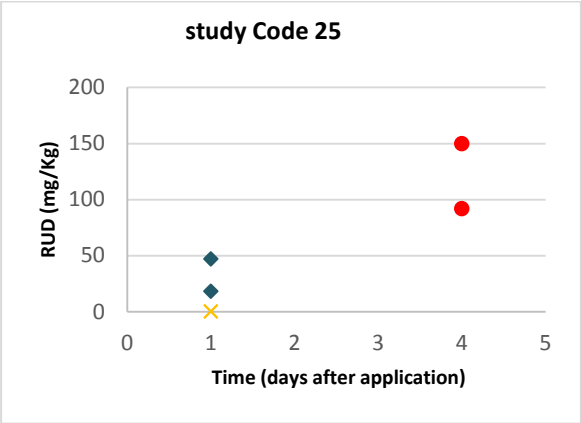


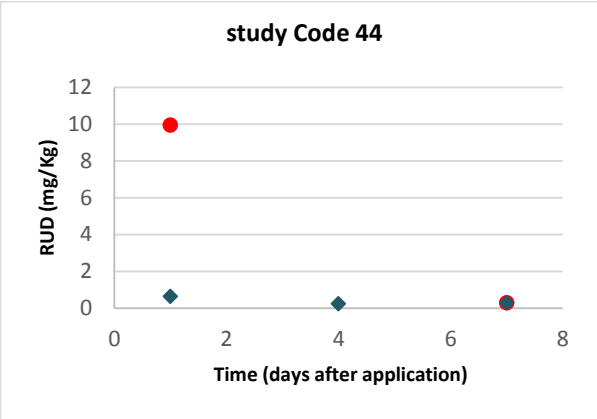


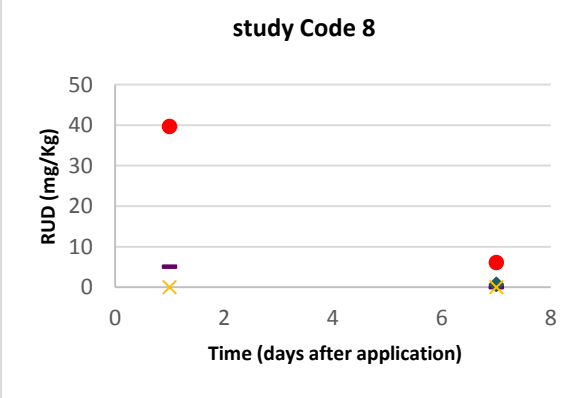


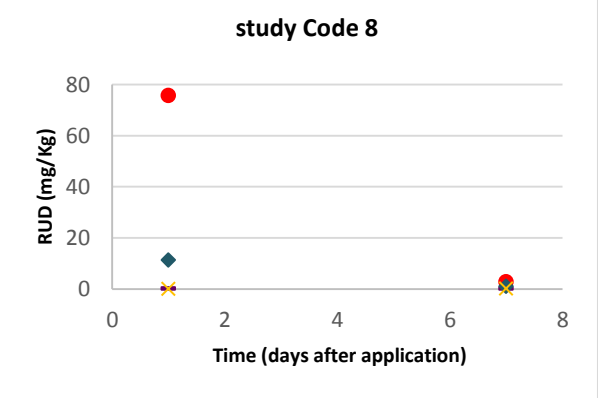


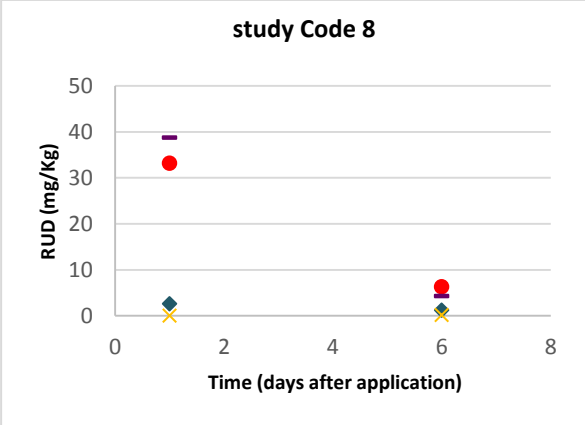


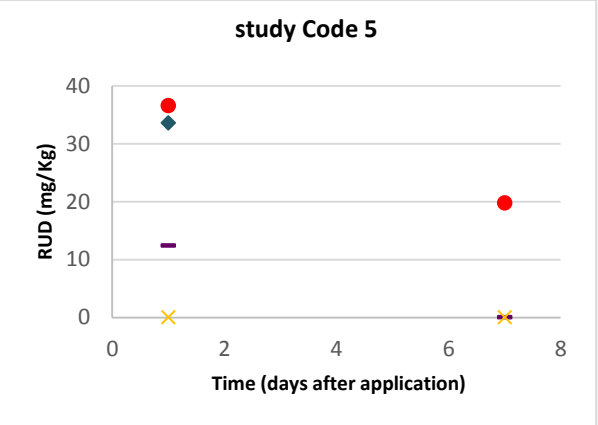


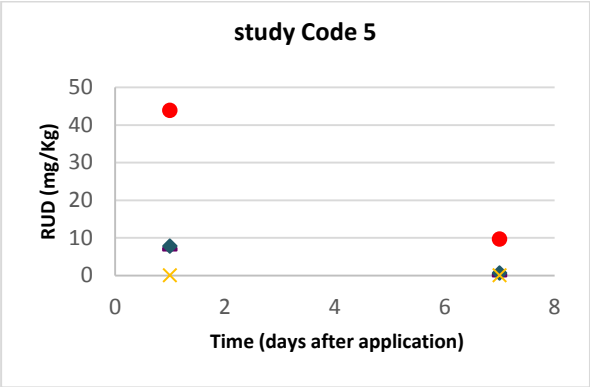


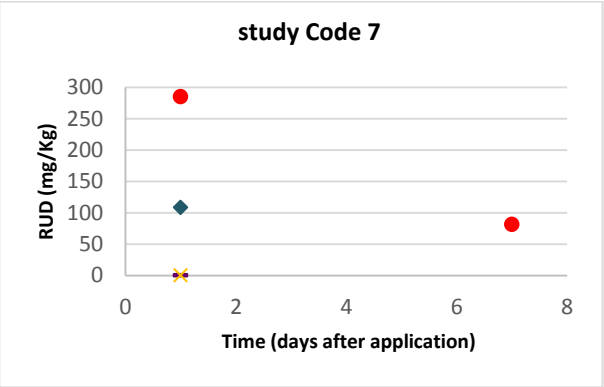


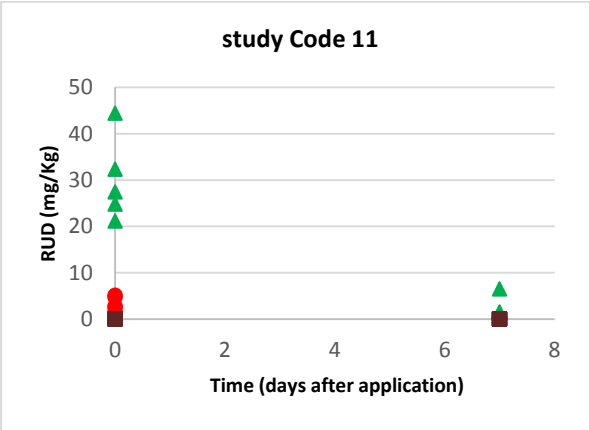


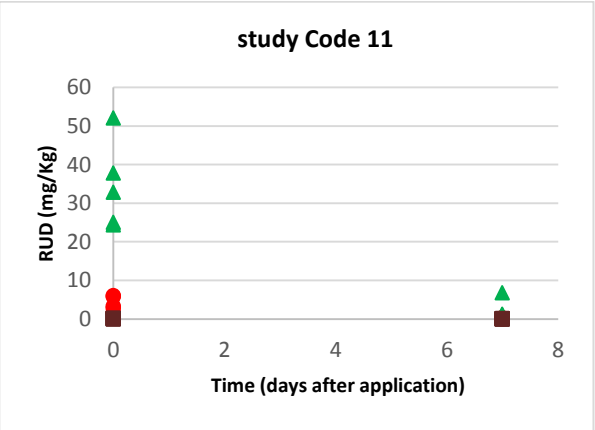


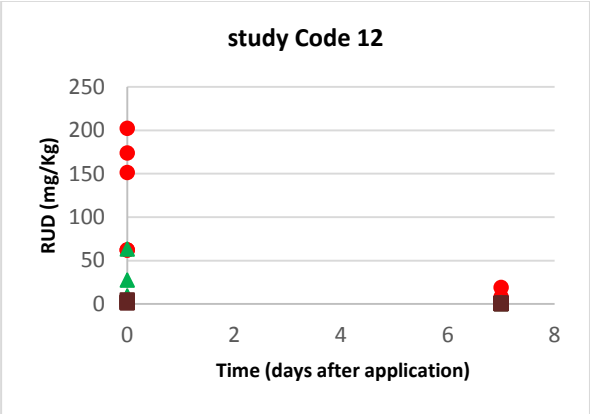


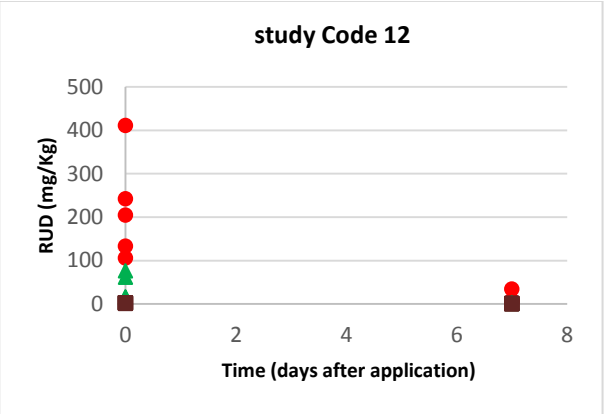












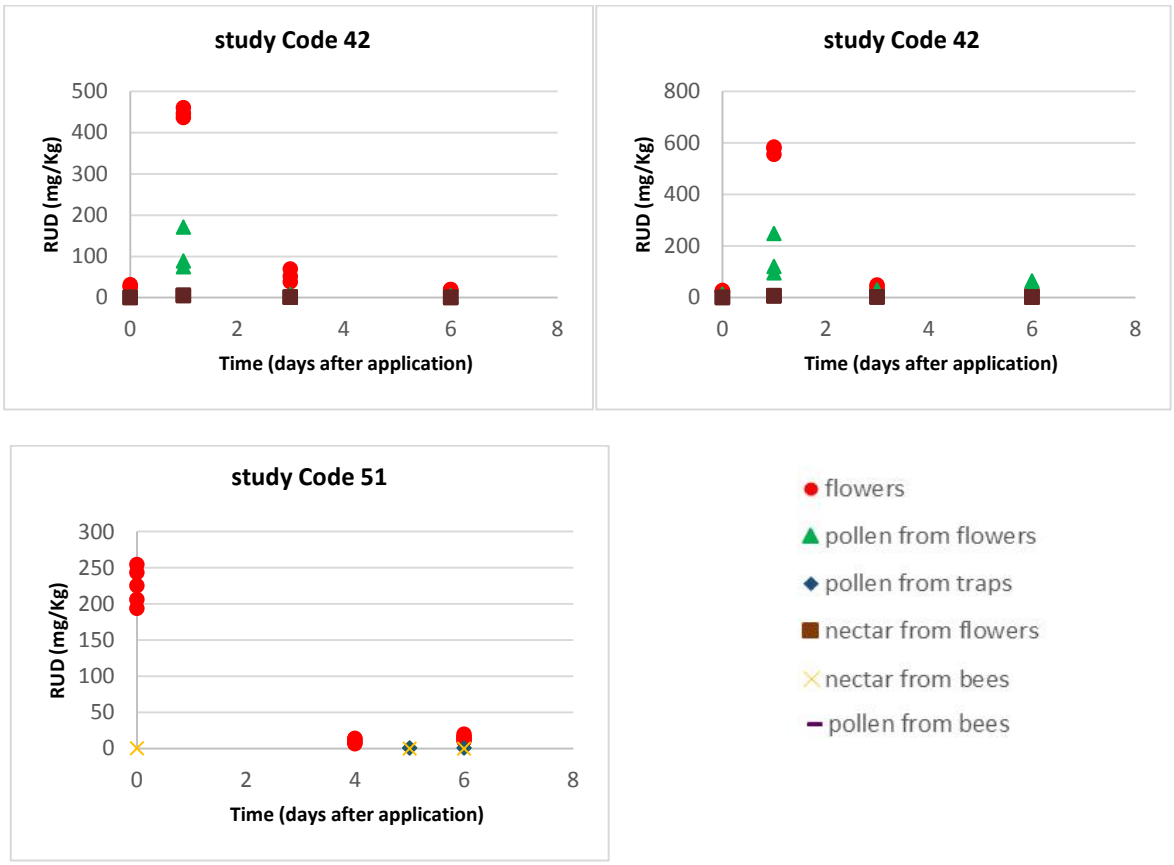


Figure 8: RUD values in flower, nectar (from bees or/and flowers) and pollen (from bees, traps and/or flowers) per study versus time in days after application.

As illustrated in the above graphs, in most studies the RUD values in whole flowers were higher than the respective values in pollen and nectar within each study. The next higher values were observed in pollen samples collected from flowers. Only in three studies (i.e. study Codes 108, 115 and 11) the RUD values in pollen collected from flowers were higher than the respective values from flowers.

3.5.2. DT_{50} & DT_{90} values

The degradation kinetics assessment for the calculation of DT_{50} / DT_{90} values was performed following the appropriate FOCUS degradation kinetics, 2014 version 1.1 flowchart (for persistence endpoints). Kinetic modelling was carried out using CAKE v 3.1 (CAKE 2015). The raw data for the calculation of DT_{50} and DT_{90} values are presented in Appendix – H.

Although DT_{50} / DT_{90} values were calculated for each collected sample included in the database, where sufficient number of sampling time points and adequate residue values above LOQ/LOD are available as described above, only some of these values were considered appropriate for further use in data analysis. Following the discussion carried out during the final meeting of the project (Dec 2016), a scoring system was developed for the allocation of each DT_{50} value into different categories.

Therefore, each one of the calculated DT_{50} values, derived from studies with 3 or more sampling points at and after application, was allocated in one of the three categories established for the

characterization of the results regarding the reliability of calculated DT_{50} values. The three (3) categories, along with the parameters/elements that have been considered, are presented below.

Category I: Fully reliable values

1. Minimum 4 sampling time points and
2. 1st sampling point should be on 0 or 1DAA and
3. Visual assessment: excellent/very good/ good and
4. χ^2<15%

Category II: Reliable with restrictions

1. Minimum 4 sampling time points and
2. 1st sampling point should be on 0 or 1DAA and
3. Visual assessment: acceptable and
4. χ^2 could be higher than 15%

Category III: Not reliable

1. Number of sampling timepoints < 4 or
2. 1st sampling after Day 1 (≥2DAA) or
3. DT_{50} values derived by "not reliable" studies

The DT_{50} values from category I and II were used for data analysis. On the contrary, "not reliable" DT_{50} values (Cat. III) were not further used for data analysis and they were considered only as supplementary data. The reliability as well as the justification for the allocation of each DT_{50} value in different reliability category was included in the worksheet "*CALCULATED_ DT_{50}_DT_{90}*" of the Database.

The Figure 9 presents the calculated DT_{50} values in pollen and nectar for each active substance. As illustrated in the graph below, the calculated DT_{50} values were in most of the cases less than 2 days for both matrices (pollen and nectar). Only in some cases with FOMC kinetic fit, the DT_{50} value in pollen was estimated to be approximately 4 days. Nevertheless, these values are much lower than the default value of 10 days that already is used according to the EFSA GD [EFSA Journal 2013:11(7): 3295].

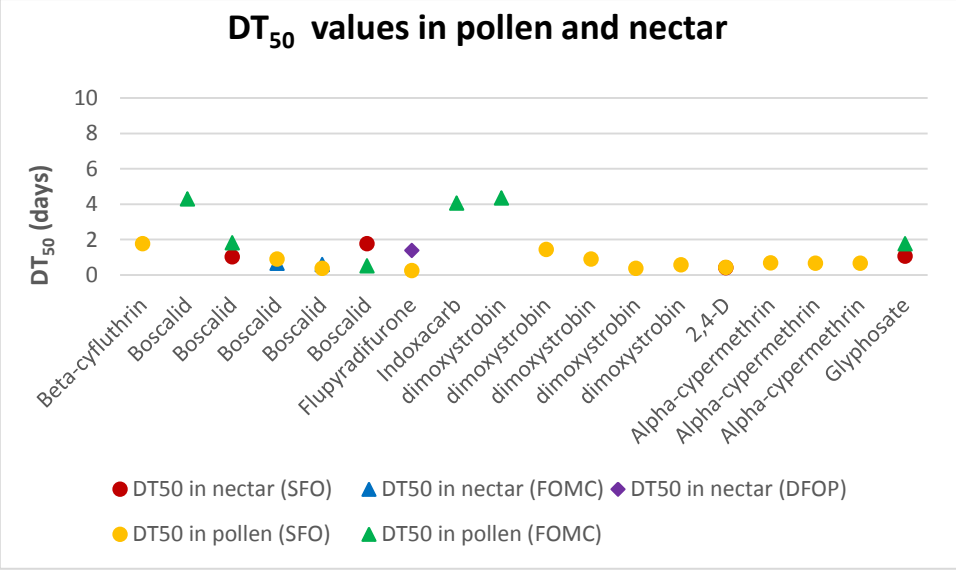


Figure 9: DT_{50} values in pollen and nectar with different types of kinetic fit (SFO, FOMC, DFOP).

3.5.3. Landscape dilution factor (LDF)

In studies where residue data of pollen and nectar from forager bees entering the hive and residue data of pollen and nectar collected directly from crops were available, the dilution factor was calculated. The LDF was calculated by using the following equation:

$$LDF\ (pollen) = \frac{RUD\ from\ pollen\ collected\ from\ bees\ or\ traps}{RUD\ from\ pollen\ collected\ from\ flowers}$$

and

$$LDF\ (nectar) = \frac{RUD\ from\ nectar\ collected\ from\ bees}{RUD\ from\ nectar\ collected\ from\ flowers}$$

Only one study i.e. Coded as Study 115 was suitable to be used for LDF calculation and the calculations that were carried out are presented in the following excel file "LDF_Code 115_OC EFSA PRAS 2015_08".

4. Data analysis results

As depicted in Section 2.5, the data analysis is focused on four core objectives. Namely, to investigate potential correlations between:

a) residue levels in pollen or nectar versus residue levels in plant foliage;

b) residue levels in pollen versus residue levels in nectar;

c) residue levels in pollen or nectar versus physical-chemical properties of the active substance (S, logPow, Koc);

d) residue decline in pollen or nectar versus residue decline in other environmental matrices.

The analysis is performed on three levels; first we examine the residue values in pollen or nectar and plant foliage as well as the residue values in pollen versus the residue values in nectar. Subsequently the residue levels in pollen and nectar vs the physical-chemical properties of the active substance are examined, followed then by an analysis of the residue decline values in nectar and pollen upon environmental characteristics.

4.1. Residue levels comparison

The first focal point of this chapter concerns the first of two core objectives of the data analysis, that is the comparison between the residue levels (RUD) in pollen and nectar and the residue levels in plant foliage.

We tackle this first by examining potential correlations between RUD in pollen and plant foliage, and RUD in nectar and plant foliage. Further, we examine potential correlations between the RUD values in pollen and the RUD values in nectar. Our aim is to examine whether the residue levels in pollen are

higher than the corresponding residue levels in nectar. In addition, we examine the effects in the residue in nectar, pollen and foliage of several attributes, such as the sampling matrix, substance and type of crop.

4.1.1. Comparison of residue levels in nectar, pollen and plant foliage

To obtain a visual indication of the residue levels we first conduct simultaneous boxplots of the residue levels (RUD) in nectar, pollen and plant foliage; see Figure 10. The plot gives a visual indication that the residue values in pollen are allegedly higher than the corresponding residue values in nectar. The lowest residue is observed in nectar; the residue in plant foliage is noticed to be higher than nectar and lower than pollen. Further, residue values in pollen display a heavier tail and greater dispersion.

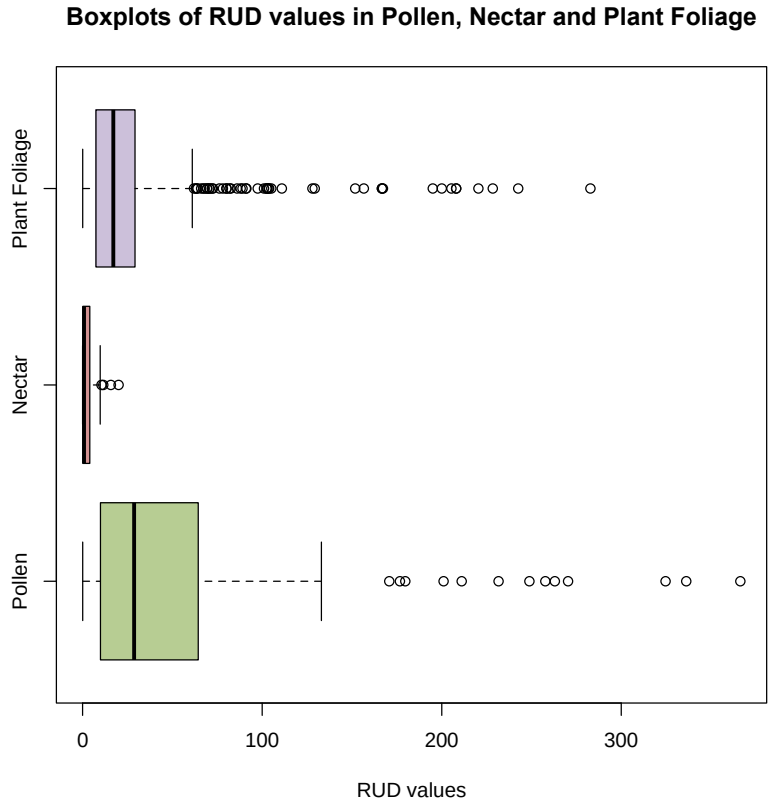


Figure 10: Boxplots of residue values in Pollen, Nectar and Plant Foliage.

Table 11 below gives a summary of the RUD values in nectar, pollen and plant foliage. In particular, the table gives the minimum, maximum, mean, median, first and third quartiles and the standard deviation of the residue values. The table also gives the number of observations available upon which the calculations of the summary statistics have been conducted.

Table 11: Summary statistics of residue levels in pollen, nectar and plant foliage.

RUD in	Min.	1st Quartile	Median	Mean	3rd Quartile	Max.	St. Dev.	# of obs.
Nectar	0.00006	0.103	0.730	2.543	3.989	20.000	3.832	75

RUD in	Min.	1st Quartile	Median	Mean	3rd Quartile	Max.	St. Dev.	# of obs.
Pollen	0.004	10.430	28.620	65.060	63.700	366.300	89.421	80
Plant Foliage	0.010	7.428	17.100	24.790	29.170	282.800	31.940	752

The summary statistics presented in Table 11 also suggest that the highest RUD values are extracted from pollen. The RUD values in nectar are significantly lower than both pollen and foliage. Furthermore, we observe that the mean RUD in pollen, nectar and foliage exceeds the median RUD, a fact that supports the observation of the distribution of the RUDs in pollen, nectar and foliage being positively skewed.

We additionally provide individual boxplots so as to better observe the variation of the residue in nectar, pollen and plant foliage separately; see Figure 11. We observe that the residue in nectar and pollen indicate an asymmetric distribution, and in particular a positive skewness as both plots display a long tail extending to the right. The residue values in plant foliage also exhibit an asymmetrical distribution. We also note that the number of observations available is significantly larger in foliage (752) than nectar (74) and pollen (80). The top plot of Figure 11 gives the RUD values in nectar, from which we note that the majority of points take values below 4. On the contrary, the middle plot gives the boxplot of the RUD values in pollen where the majority of the observations take values up to 60. The bottom plot gives the RUD values in foliage, where we observe that the majority of values are below 30. All boxplots signify that outliers are detected in all three datasets.

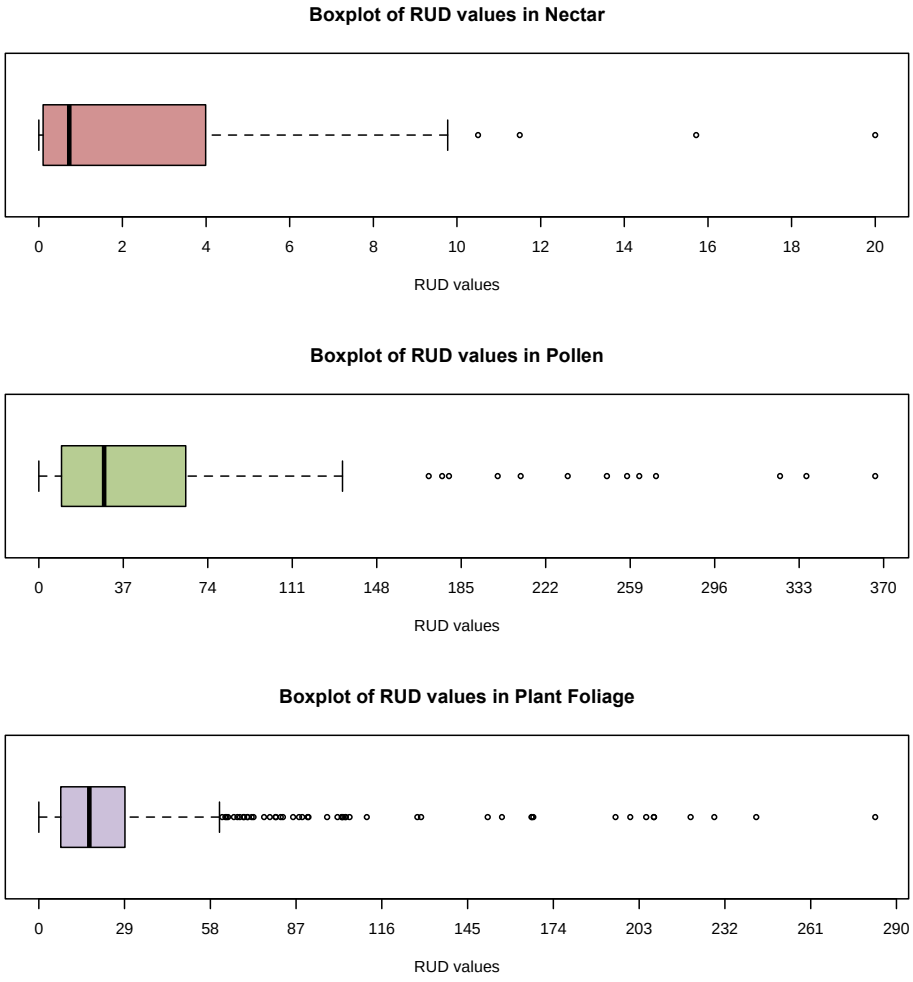


Figure 11: Separate boxplots of the residue values in nectar, pollen and plant foliage.

Location comparison

In order to compare the location of the residue values in nectar, pollen and plant foliage, we employ the t-test of mean comparison. The t-test is a hypothesis test applied in order to determine whether the difference in the values of two variables is significantly different or greater than zero, by comparing their means. Before conducting the t-test, we firstly perform an F-test to compare the variances of the two variables, so as to take into account equality or inequality of variances when conducting the t-test.

Table 12 presents the results of the t-tests testing the null hypotheses of equality in means to the alternative hypothesis that the mean RUD of sample 1 is greater than the mean RUD of sample 2. In all three tests the null hypothesis is rejected, therefore we result that the residue in Pollen is higher than the residue in Nectar (top row), the residue in Plant Foliage is also higher than the residue in Nectar (middle row), and lastly, that the residue Pollen is higher than the residue in Plant foliage (bottom row).

Table 12: T-test for mean comparison on RUD values in nectar, pollen and plant foliage.

	F-test to compare variances	Welch two sample t-test

	Ratio of variances	p-value	t-statistic	Mean RUD sample 1	Mean RUD sample 2	p-value
Pollen vs Nectar	544.550	<0.001	6.247	65.062	2.543	<0.001
Plant Foliage vs Nectar	69.474	<0.001	17.855	24.789	2.543	<0.001
Pollen vs Plant Foliage	7.838	<0.001	4.001	65.062	24.789	<0.001

Table 13 shows the results of the t-tests comparing the mean residue in pollen, nectar, pollen and nectar from flower, pollen and nectar from bees, and pollen from traps (sample 1) with the mean residue in plant foliage. The alternative hypothesis is that the mean residue in nectar/pollen is different than the mean residue in plant foliage, while the null hypothesis is that the two means are equal.

Table 13: T-tests for mean comparison of RUD values in nectar and pollen with the RUD values in plant foliage.

	F-test to compare variances		Welch two sample t-test			
RUD in	**Ratio of variances**	**p-value**	**t-statistic**	**Mean RUD of sample 1**	**Mean RUD of foliage**	**p-value**
Pollen vs Plant Foliage	7.838	<0.001	4.001	65.062	24.789	<0.001
Nectar vs Plant Foliage	0.014	<0.001	-17.855	2.543		<0.001
Pollen from flower vs Plant Foliage	11.616	<0.001	4.098	101.436		<0.001
Nectar from flower vs Plant Foliage	0.018	<0.001	-15.581	3.295		<0.001
Nectar from bees vs Plant Foliage	0.012	<0.001	-17.443	2.158		<0.001
Pollen from bees vs Plant Foliage	4.204	<0.001	0.913	36.799		0.370
Pollen from traps vs Plant Foliage	2.918	<0.001	2.033	39.506		0.042

The F-test to compare variances rejects the null hypothesis of equality of variances in all cases.

The t-tests reject the null hypothesis of equality of the mean residue in pollen, nectar, pollen and nectar from flower, nectar from bees and pollen from traps with the mean residue in plant foliage, while the null hypothesis is not rejected between pollen from bees and plant foliage, suggesting that the equality of mean residue between pollen from bees and plant foliage cannot be rejected.

As exhibited, significant differences in mean residue are detected between plant foliage and pollen, pollen from flower, nectar from flower, nectar from bees and pollen from traps. The highest difference is noticed between plant foliage and pollen from flower; mean difference 76.65, indicating that the residue in pollen from flower exceeds the residue in plant foliage on average by 76.65.

Last, Figure 12 shows boxplots of the residue values in plant foliage, pollen and nectar for all sampling matrices, and the plots amplify the outcomes already resulted from the t-tests.

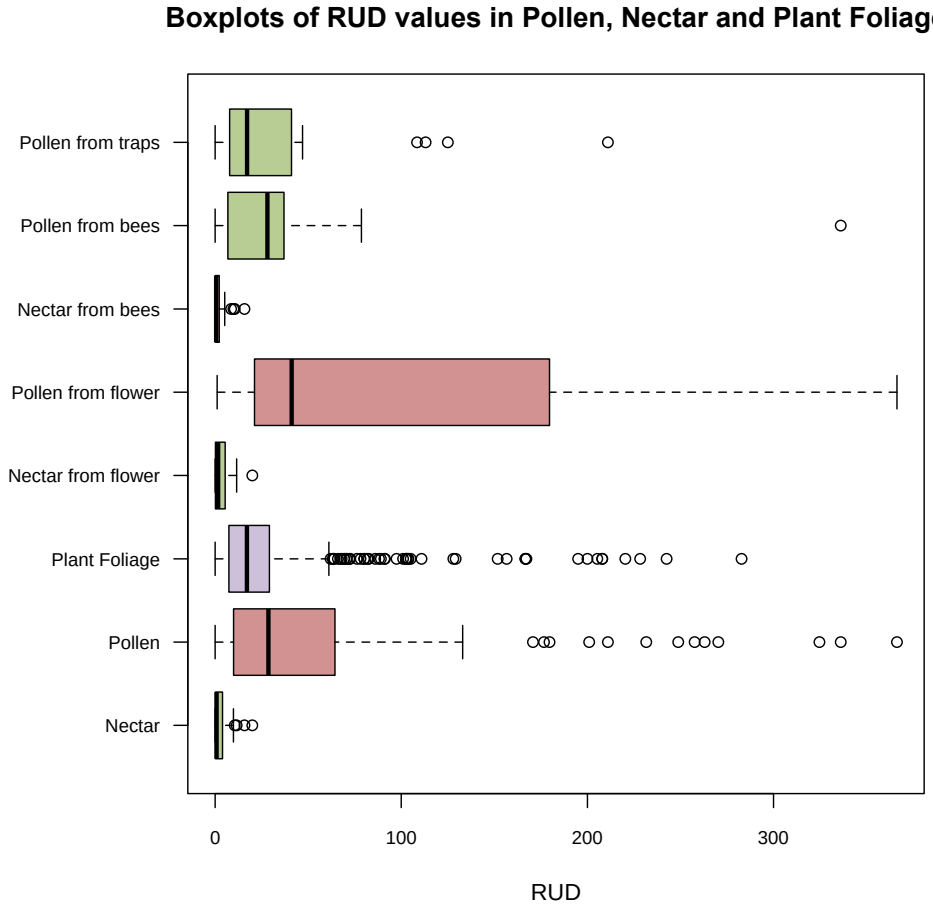


Figure 12: Boxplots of the residue values in pollen, nectar and plant foliage.

In conclusion:

- The residue values in nectar are significantly lower than the residue values in both pollen and plant foliage.
- The residue values in pollen are the highest.
- The residue values in plant foliage are higher than the residue in nectar.
- The distribution of the residue values in nectar, pollen and plant foliage is asymmetrical, and in particular, positively skewed.
- Statistically significant differences are detected between plant foliage and pollen and plant foliage and nectar. In particular, mean residue in pollen is higher than the mean residue in plant foliage, and mean residue in plant foliage is higher than the mean residue in nectar.
- The highest difference is detected between pollen from flower and plant foliage.

- There do not exist statistically significant differences between pollen from bees or traps with plant foliage.

4.1.2. RUD in pollen and nectar - Sampling matrix related differences

In this section we examine possible differences in the RUD values in nectar and pollen that arise due to the different sampling matrices from which the residue is drawn – flower/bees/traps.

Table 14 displays a summary of the RUD values in nectar and pollen for the different sampling matrices.

Table 14: Summary statistics of residue levels in pollen and nectar for each sampling matrix.

Sampling Matrix	Min.	1st Quartile	Median	Mean	3rd Quartile	Max.	St. Dev.	# of obs.
Nectar from flower	0.010	0.148	1.626	3.295	5.361	20.000	4.246	33
Pollen from flower	1.071	21.960	41.130	101.400	178.900	366.300	108.856	34
Nectar from bees	0.00006	0.185	0.549	2.158	2.124	15.720	3.524	38
Pollen from bees	0.004	6.857	28.040	36.800	36.950	336.200	65.486	25
Pollen from traps	0.009	7.775	17.130	39.510	41.000	211.100	54.556	21

The findings displayed in the table above suggest higher values of residue in pollen than nectar, regardless of the sampling matrix. Additionally, we detect that the RUD values in both nectar and pollen are the largest when the residue measurements are extracted from flower.

Figure 13 presents boxplots of the residue values in pollen and nectar for the five types of sampling matrices. The figure amplifies the results described above. It is understandable that the RUD values in pollen are higher than the RUD values in nectar, and this is even more acute in the case of RUD values from flowers.

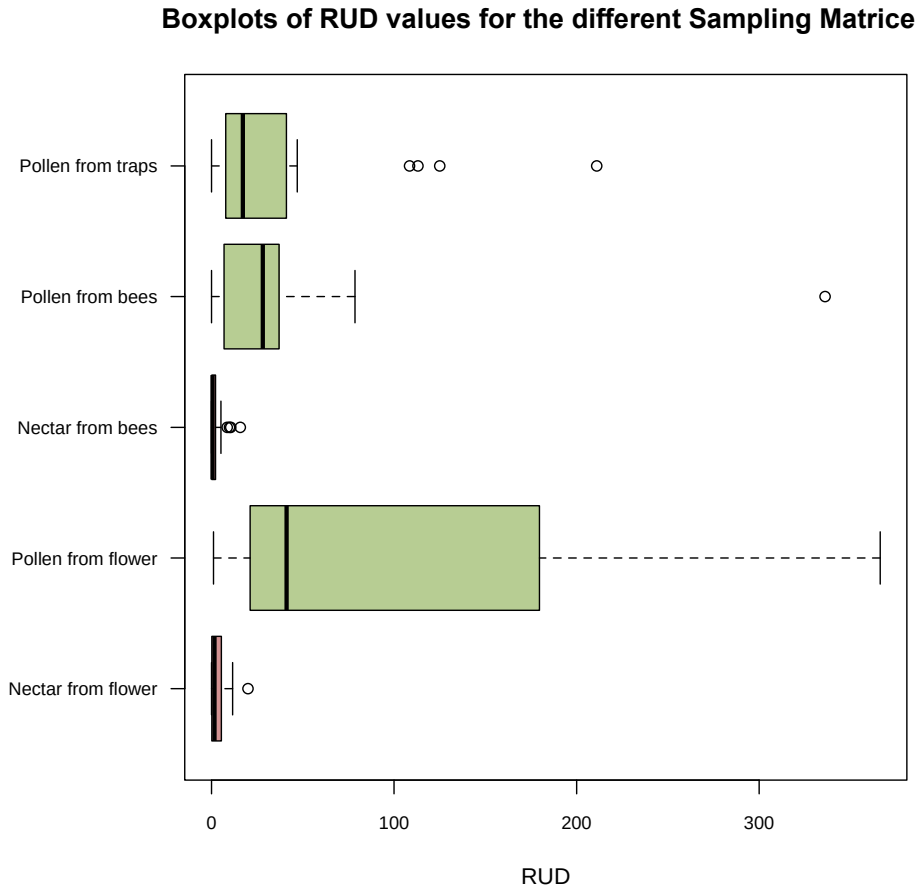


Figure 13: Boxplots of the RUD values for the different sampling matrices.

Analysis of variance

In addition, we perform an analysis of variance to investigate possible differences between the sampling matrices. The aim of the analysis of variance is to examine if the sampling matrix has any effect on the mean (or median) residue values.

Figure 14 gives graphically the effects of the sampling matrix on the mean and median RUD values. The horizontal line gives the mean (or median) residue value of all data points, and the sampling matrices are marked in each plot by a smaller horizontal segment corresponding to the mean (or median) residue value in each sampling matrix. The left plot is formed based on the mean and the right plot is formed based on the median. The reason we compare both measures of location is because the distribution of the residue values is asymmetrical.

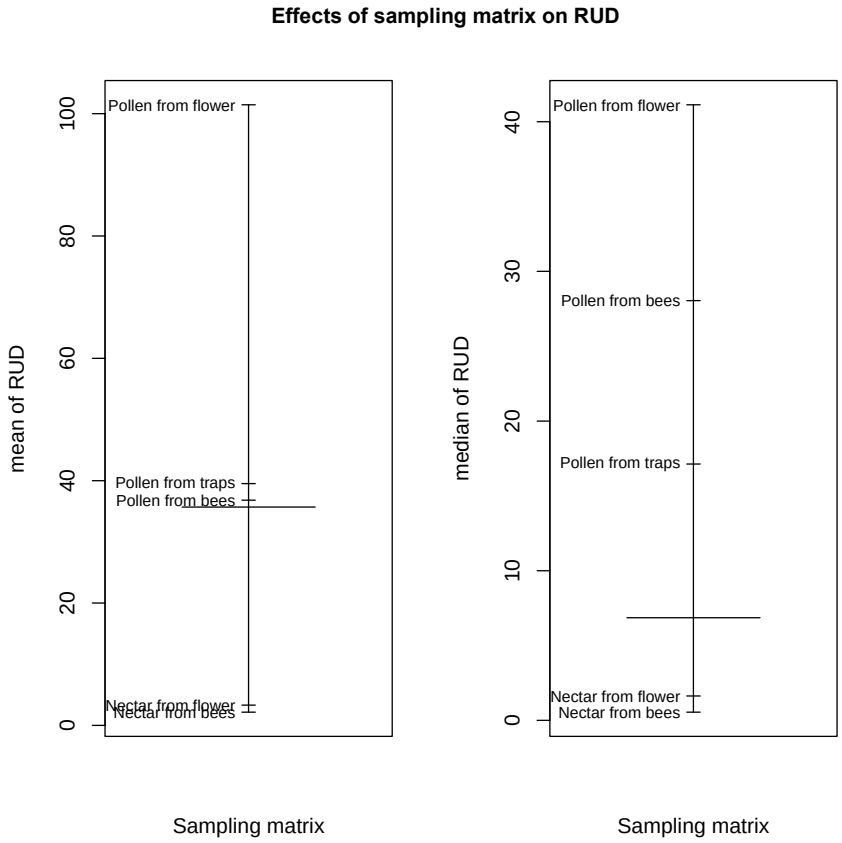


Figure 14: Effects of the sampling matrix on the residue levels in nectar and pollen.

In particular, pollen from bees is found to be in the same level group with pollen from traps in terms of the mean residue values, whereas this is not the case in terms of median residue values. We note two categories formed by pollen from bees and pollen from traps on the one hand, and nectar from bees and nectar from flower on the other hand. Additionally, RUD values in pollen are higher than RUD values in nectar, and finally the largest RUD values are measured in flowers.

Table 15, gives an analysis of variance (ANOVA) of the residue values on the type of sampling matrix. Analysis of variance is the partition of the observed variance of a variable – in our case the variance of the residue values – to the levels of a factor. Our aim is to analyse the variability of the residue values to the various sampling matrices. The null hypothesis of the ANOVA test is that the population means of the Sampling matrices are equal, that is the analysis of variance provides a test of equality in the mean residue values between the sampling matrices.

Table 15: Analysis of variance (ANOVA) table of the residue values on the type of sampling matrix (excluding the extreme measurements).

Analysis of variance (ANOVA) table

	Degrees of freedom	Sum of Squares	Mean Sum of Squares	F-statistic	p-value
Sampling Matrix	4	224661	56165	14.79	<0.001
Residuals	146	554523	3798		

Analysis of variance (ANOVA) table					
	Degrees of freedom	**Sum of Squares**	**Mean Sum of Squares**	**F-statistic**	**p-value**
Kruskal-Wallis Rank Sum Test					
Test statistic		**Degrees of freedom**		**p-value**	
84.574		4		<0.001	

We note however that the results presented in Table 14 and Figure 13 suggest that the normality assumption of the ANOVA test is violated. Furthermore, the assumption of homogeneity needs to be verified; to validate this we run a Levene's test of homogeneity of variances, which is found to reject the null hypothesis of homogeneity of variances (p-value<0.001). However, even though the assumptions of normality and homogeneity are violated, the one-way ANOVA can tolerate deviations from the normality assumption with a small effect on the probability of a false rejection of the null hypothesis (Type I error) and an approach that provides a protection against non-normality and heterogeneity is to perform a non-parametric test. Therefore, in addition to the one-way ANOVA test we also perform the non-parametric Kruskall-Wallis rank sum test which tests the null hypothesis that the residue values from all sampling matrices come from the same population; see Table 15.

Examining the results presented in Table 15, we note that the p-value of the analysis of variance test, being <0.001, suggests that the null hypothesis is rejected, **which means that the mean residue values are different between the various sampling matrices**. The same results are identified from the non-parametric test.

Seeing that a difference in the sampling matrices as regards the residue values has been identified, we further wish to identify these differences, that is we want to identify which sampling matrices are significantly different. This is accomplished by employing post hoc tests for pairwise multiple comparisons. We consider the multiple comparisons method of Tukey – which is based on the analysis of variance. Accordingly, the 95% confidence intervals of the mean RUD values between all pairs of sampling matrices are computed. The idea is that confidence intervals which include 0 suggest that the corresponding sampling matrices null hypothesis is not rejected. The results are presented in Table 16.

Table 16: Tukey's multiple comparisons of the mean residue values between the sampling matrices.

Sampling matrices pairs	**Tukey's Multiple comparisons**		
	Mean difference	**95% Confidence Interval**	**p-value**
Nectar from flower - Nectar from bees	1.137	(-39.369,41.642)	0.999
Pollen from bees - Nectar from bees	34.640	(-9.196,78.478)	0.192
Pollen from flower - Nectar from bees	99.278	(59.092,139.463)	0
Pollen from traps - Nectar from bees	37.348	(-8.939,83.635)	0.175
Pollen from bees - Nectar from flower	33.504	(-11.632,78.640)	0.248

Sampling matrices pairs	Tukey's Multiple comparisons		
	Mean difference	95% Confidence Interval	p-value
Pollen from flower - Nectar from flower	98.141	(56.543,139.739)	0
Pollen from traps - Nectar from flower	36.211	(-11.308,83.729)	0.224
Pollen from flower - Pollen from bees	64.637	(19.788,109.486)	0.001
Pollen from traps - Pollen from bees	2.707	(-47.682,53.095)	0.999
Pollen from traps - Pollen from flower	-61.930	(-109.176,-14.684)	0.004

Figure 15 provides graphical representations of the Tukey's multiple comparisons 95% confidence intervals given in Table 16, in the same order as presented on the table. In the cases of (1) nectar from flower-nectar from bees, (2) pollen and nectar from bees, (3) pollen from traps and nectar from bees, (4) pollen from bees and nectar from flower, (5) pollen from traps and nectar from flower and (6) pollen from traps and pollen from bees, the results indicate that there do not exist significant differences in the mean residue values. On the contrary, for the cases of (1) pollen from flower and nectar from bees, (2) pollen from flower and nectar from flower, (3) pollen from flower and pollen from bees, and (4) pollen from traps and pollen from flower there exist significant differences in the mean RUD values.

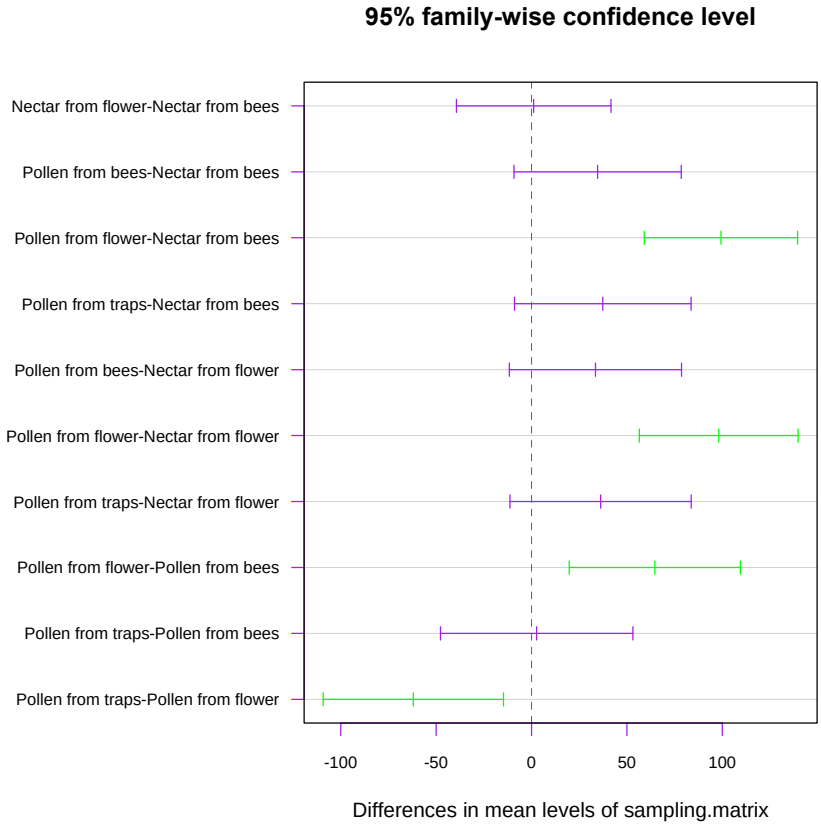


Figure 15: Tukey's multiple comparisons 95% confidence intervals of the difference in the mean RUD for all pairs of sampling matrices

Tests of association

Table 17 presents the results of tests of association between nectar and pollen for the various types of sampling matrices. Our aim is to examine possible connections between nectar and pollen, that is whether positive or negative correlation exists between the two. In all hypothesis tests given in the table below, the null hypothesis is that the true correlation is zero, and the alternative hypothesis is that the true correlation is greater than 0. The tests are conducted based on complete pairs of observations, that is we do not include measurements in which either only nectar or pollen was measured. The correlation analysis is conducted by employing Pearson and Spearman correlation tests.

Table 17: Correlation tests between nectar and pollen.

Test for association/correlation between nectar and pollen					
	Pearson			Spearman	
	Correlation coefficient	95% Confidence Interval	p-value	Correlation coefficient	p-value
Nectar vs Pollen	0.796	(0.711,1)	<0.001	0.580	<0.001

Test for association/correlation between nectar and pollen					
	Pearson			Spearman	
	Correlation coefficient	95% Confidence Interval	p-value	Correlation coefficient	p-value
Nectar from flower vs Pollen from flower	0.844	(0.733,1)	<0.001	0.731	<0.001
Nectar from bees vs Pollen from bees	0.950	(0.894,1)	<0.001	0.804	<0.001
Nectar from bees vs Pollen from traps	0.452	(0.048,1)	0.034	-0.057	0.586

In general, the Spearman correlation coefficient is more robust against outliers. This is because the Spearman correlation coefficient is a non-parametric measure of association that does not rely on the underlying distribution of the data points and depends on the median instead of the mean.

The results suggest the existence of **strong positive correlation between RUD values in nectar and pollen for all sampling matrices; except for nectar from bees with pollen from traps, with the largest correlation being detected when nectar and pollen are extracted directly form the bees**. The p-value in all tests apart from the test on nectar from bees versus pollen from traps, is significantly small, therefore the null hypothesis is rejected in these cases.

Location Comparison

In the following, we perform t-tests between nectar and pollen. The alternative hypothesis in all t-tests displayed in Table 18 is that the mean RUD value in pollen is greater than the mean RUD value in nectar. The null hypothesis of all t-tests is rejected in all tests at 5% significance level, suggesting that **the mean RUD values in pollen are indeed greater than the corresponding mean RUD values in nectar**. The same results arise when conducting the non-parametric Mann-Whitney test for location. In particular, the highest mean difference in residue is detected between nectar and pollen from flower (mean difference 98.141), followed by the difference between nectar and pollen (mean difference 62.518).

Table 18: T-tests for mean comparison of RUD values in nectar and pollen for the different types of sampling matrices.

	F-test to compare variances		Welch two sample t-test				Mann-Whitney test
	Ratio of variances	p-value	t-statistic	Mean RUD pollen	Mean RUD nectar	p-value	p-value
Nectar vs Pollen	544.550	<0.001	6.247	65.062	2.544	<0.001	<0.001
Nectar from flower vs Pollen from flower	657.198	<0.001	5.253	101.436	3.295	<0.001	<0.001

	F-test to compare variances		Welch two sample t-test				Mann-Whitney test
	Ratio of variances	p-value	t-statistic	Mean RUD pollen	Mean RUD nectar	p-value	p-value
Nectar from bees vs Pollen from bees	345.318	<0.001	2.642	36.799	2.158	0.007	<0.001
Nectar from bees vs Pollen from traps	239.666	<0.001	3.134	39.506	2.158	0.003	<0.001

Regression analysis

Linear regression results between RUD values in nectar and pollen are displayed in Tables 19 and 20 below. In particular, we perform linear regression in order to model the relationship between RUD values in nectar and pollen for all sampling matrices. The model we examine is

$$Nectar_{RUD} = a + bPollen_{RUD}$$

where a is the intercept and b is the change in the RUD in nectar caused by a unit change in the RUD in pollen. Table 19 gives the estimated coefficients $\hat{a}$ and $\hat{b}$ of the regression along with their standard errors, the corresponding t-statistic and p-value, and also the coefficient of determination R^2 and the residual standard error.

In all sampling matrices, the findings of the regression support the findings from the correlation analysis. In particular, we observe a strong relationship between nectar and pollen, and this is an even stronger relationship in the case where nectar and pollen are extracted from bees, as indicated by the value of the coefficient of determination. Recall that the coefficient of determination gives the percentage of variation which is explained by the model; in the case of sampling directly from bees we observe a percentage of nearly 90%, and in the case of sampling from flower we observe a percentage of 71%.

Table 19: Linear regression between RUD values in nectar and pollen for the different types of sampling matrices.

	Coefficients		Std. Error	t-value	p-value	R^2	Residual Std. Error
Nectar - Pollen	$\hat{a}$	0.292	0.351	0.832	0.408	0.633	2.370
	$\hat{b}$	0.033	0.003	10.988	<0.001		
Nectar from flower - Pollen from flower	$\hat{a}$	-0.124	0.561	-0.220	0.827	0.712	2.314
	$\hat{b}$	0.033	0.004	8.761	<0.001		
Nectar from bees - Pollen from bees	$\hat{a}$	0.130	0.289	0.452	0.656	0.902	1.149
	$\hat{b}$	0.048	0.004	13.195	<0.001		
Nectar from bees - Pollen from traps	$\hat{a}$	0.976	1.040	0.939	0.363	0.205	3.321
	$\hat{b}$	0.028	0.014	1.965	0.068		

The estimated regression lines are given in the following Table 20 below, where we can see that a negative intercept regression line results in the case of nectar from bees and pollen from traps.

Table 20: Estimated regression lines

	Estimated regression line
Nectar - Pollen	$\widehat{Nectar}_{RUD} = 0.292 + 0.033 Pollen_{RUD}$
Nectar from flower - Pollen from flower	$\widehat{Nectar}_{RUD} = -0.124 + 0.033 Pollen_{RUD}$
Nectar from bees - Pollen from bees	$\widehat{Nectar}_{RUD} = 0.130 + 0.048 Pollen_{RUD}$
Nectar from bees - Pollen from traps	$\widehat{Nectar}_{RUD} = 0.976 + 0.028 Pollen_{RUD}$

Lastly, Figure 16 gives graphical representations of the RUD values in nectar and pollen for the different types of sampling matrices, along with the estimated regression lines as given above.

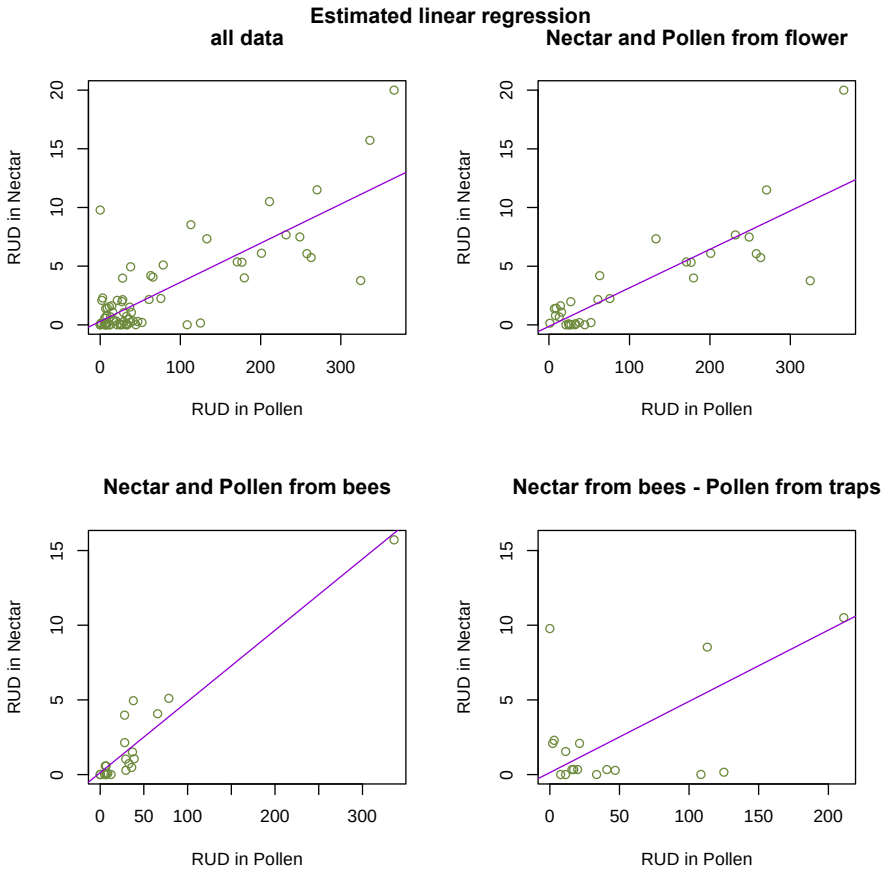


Figure 16: Estimated linear regression of Nectar-Pollen RUD for the different sampling matrices.

In conclusion:

- The residue in pollen is higher than nectar irrespective of the sampling matrix.
- There exist statistically significant differences among the sampling matrices.
- The residue in both pollen and nectar is highest when extracted from flower.
- Strong positive correlation is detected between nectar and pollen, nectar and pollen from bees, and nectar and pollen from flower; the strongest relationship is observed in the case where nectar and pollen are extracted from bees.

4.1.3. RUD in pollen and nectar - Type of crop related differences

In the following we focus on identifying possible differences in RUD values in nectar and pollen arising from the different types of crop. We consider three types of crop: *Phacelia tanacetifolia*, *Rapeseed* and all other types of crop.

Figure 17 below gives boxplots of the residue levels in pollen (left plot) and nectar (right plot) for the three different types of crop. Both plots show that the residue levels are found significantly higher in rapeseed.

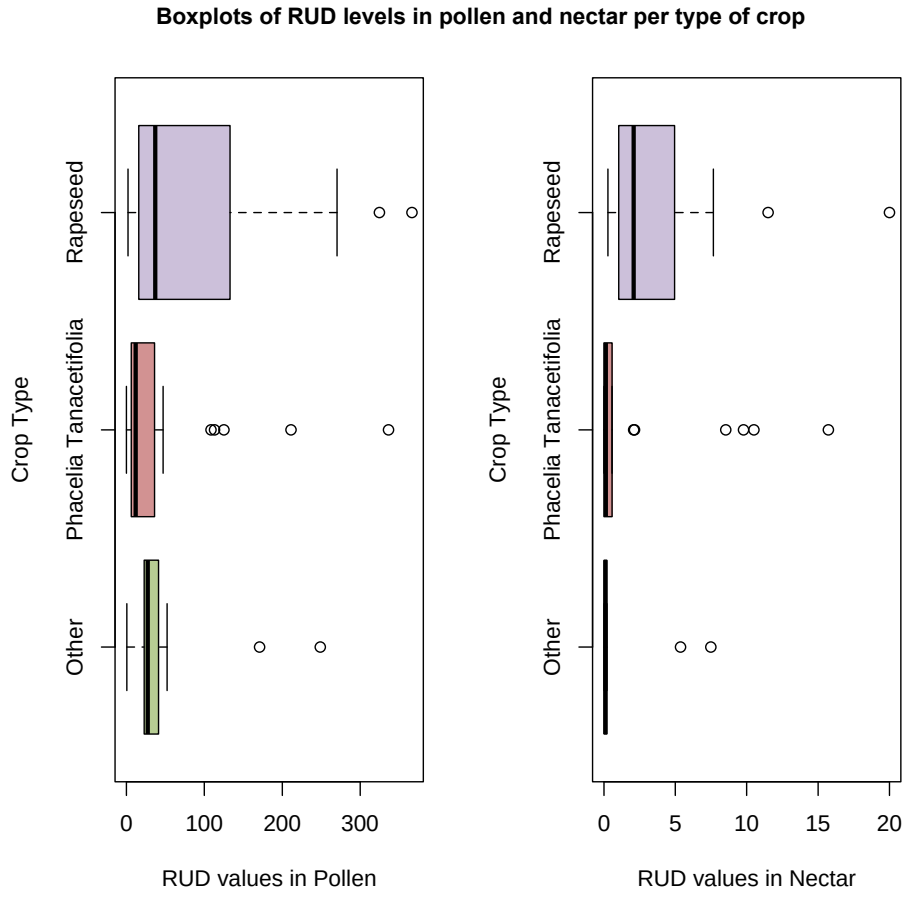


Figure 17: Boxplots of the residue levels in pollen and nectar per type of crop

Table 21 presents summary statistics of the RUD levels in pollen and nectar for the three types of crop. From the summary results we see that for all three types of crop the mean and median RUD values in pollen are higher than the corresponding values in nectar.

The results presented in the table show that the RUD levels detected in *Rapeseed* notably exceed the RUD levels detected in *Phacelia tanacetifolia* and other types of crop.

Table 21: Summary statistics of the residue levels in pollen and nectar for the different types of crop.

RUD	Min.	1st Quartile	Median	Mean	3rd Quartile	Max.	St. Dev.	# of obs.
Type of crop: *Phacelia tanacetifolia*								
Pollen	0.004	6.416	11.850	43.980	34.870	336.200	75.241	28
Nectar	0.00006	0.0007	0.125	2.064	0.571	15.720	4.231	25
Pollen from bees	0.004	5.398	6.900	33.760	22.270	336.200	84.511	15
Pollen from traps	0.009	10.940	26.320	55.260	108.500	211.100	64.439	13
Nectar from bees	0.00006	0.075	0.280	2.456	2.083	15.720	4.523	21
Type of crop: Rapeseed								
Pollen	2.083	15.900	36.950	87.060	133.000	366.300	102.800	37
Nectar	0.282	1.037	2.083	3.392	4.945	20.000	3.820	37
Pollen from flower	6.790	15.290	104.300	134.900	238.200	366.300	120.240	20
Nectar from flower	0.680	1.575	3.883	4.754	6.075	20.000	4.579	20
Pollen from bees	27.960	30.290	36.480	41.360	38.670	78.560	16.944	10
Pollen from traps	2.083	7.246	15.900	15.800	18.550	41.000	13.055	7
Nectar from bees	0.282	0.333	1.054	1.790	2.292	5.100	1.698	17
Type of crop: Other								
Pollen	0.629	22.780	27.420	50.150	41.130	248.800	67.984	15
Nectar	0.010	0.010	0.103	1.051	0.202	7.486	2.425	13
Pollen from flower	1.071	24.540	29.880	53.690	42.790	248.800	69.103	14
Nectar from flower	0.010	0.010	0.103	1.051	0.202	7.486	2.425	13

Furthermore, Figure 18 displays boxplots of the residue levels in pollen and nectar per type of crop and per sampling matrix. The plots indicate that the **residue values in pollen are higher than the corresponding values in nectar, irrespective of the sampling matrix and type of crop**. More specifically, the highest residue values are detected when the sampling matrix is pollen from flower and the type of crop is rapeseed. In addition, when considering the three types of crop separately, **residue in pollen from each of the sampling matrices is higher than the residue in nectar, for all three types of crop**.

We note that RUD values from *Phacelia tanacetifolia* when the sampling matrix is pollen/nectar from flower are not available, hence are not included in this comparison. Also, only one RUD observation is available from pollen from bees, traps and nectar from bees, for other types of crop than *Phacelia tanacetifolia* and rapeseed, therefore this is also not included.

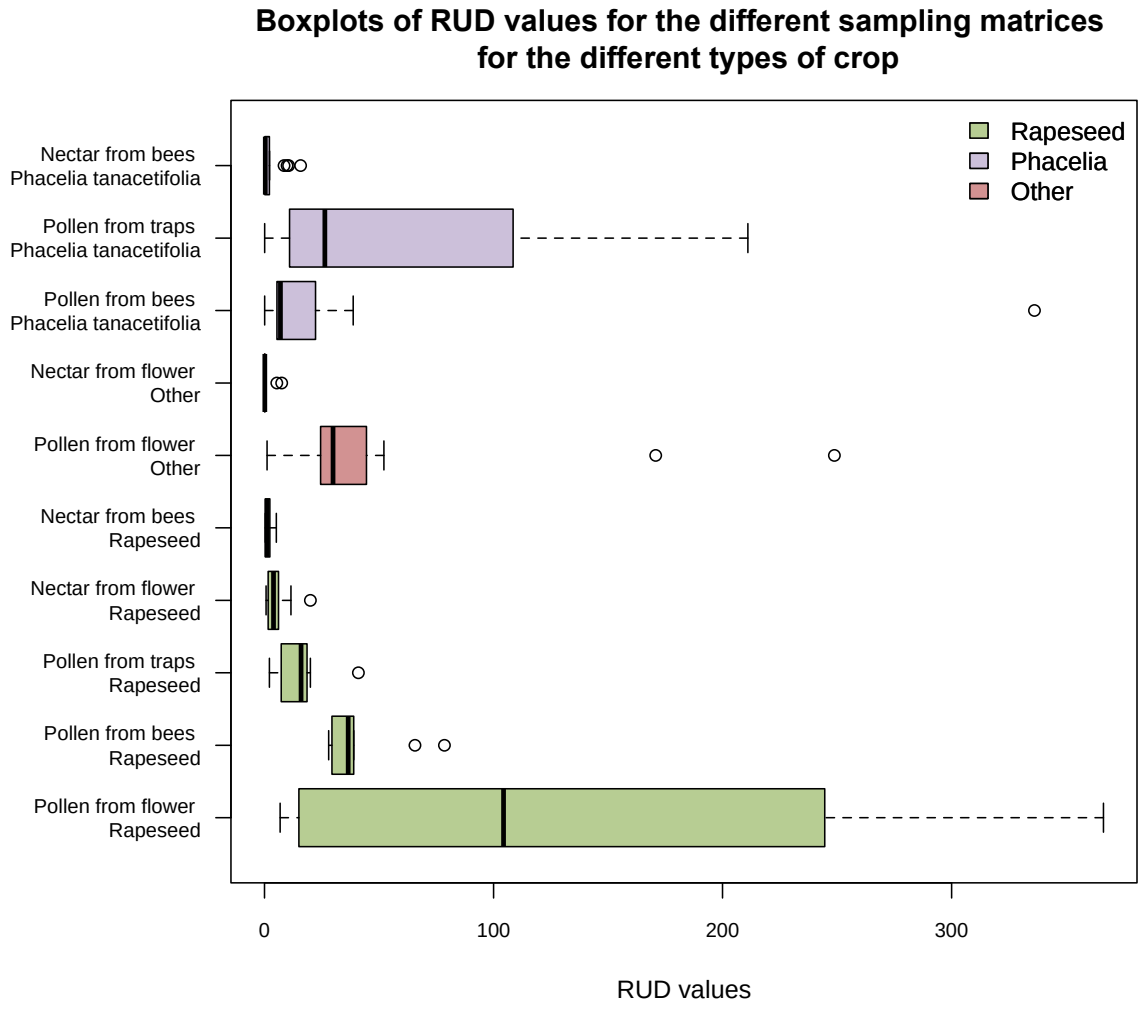


Figure 18: Boxplots of the residue levels in pollen and nectar per type of crop and per sampling matrix.

Analysis of variance

In order to explore crop related differences between the residue levels in nectar and pollen, we also perform analysis of variance. The effects of the type of crop based in the mean (left plot) and the median (right plot) are shown in Figure 19. Comparing the right plots that correspond to the median, we observe that *Rapeseed* and *Phacelia tanacetifolia* form two distinct levels, with rapeseed clearly exhibiting larger residue levels than all other crop types.

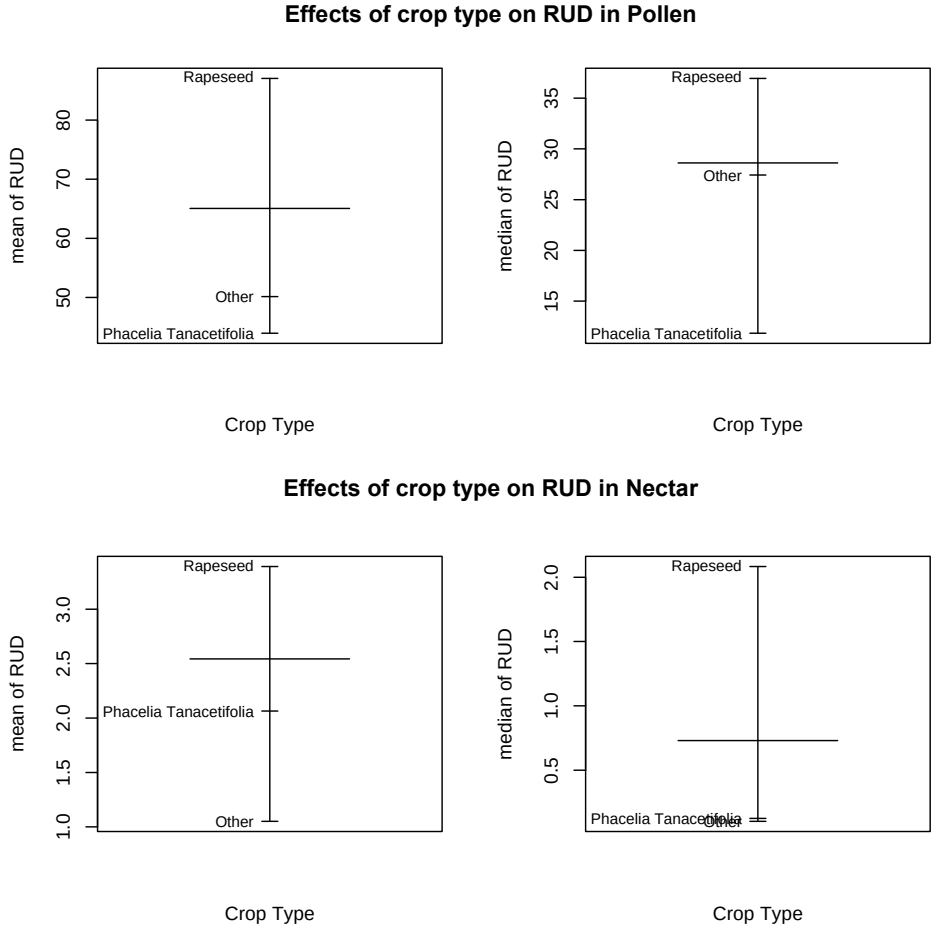


Figure 19: Effects of the type of crop on the residue levels in nectar and pollen.

Furthermore, Table 22 gives the analysis of variance of the residue levels in pollen and nectar on the type of crop. The results suggest that both in the case of pollen (p-value=0.121>0.005) and in the case of nectar (p-value=0.123>0.005), no significant differences are detected between the types of crop with a significance level of 5%. However, the non-parametric Kruskal-Wallis test is also performed as a remedy to the violation of the normality assumption, and we note that the non-parametric test detects differences between the types of crop both in the case of nectar and in the case of pollen as well.

Table 22: Analysis of variance (ANOVA) table of the residue values in pollen and nectar on the type of crop.

Analysis of variance (ANOVA) table					
	Degrees of freedom	**Sum of Squares**	**Mean Sum of Squares**	**F-statistic**	**p-value**

Analysis of variance (ANOVA) table					
	Degrees of freedom	**Sum of Squares**	**Mean Sum of Squares**	**F-statistic**	**p-value**
Pollen					
Type of crop	2	33694	16847	2.169	0.121
Residuals	77	598001	7766		
Nectar					
Type of crop	2	61.300	30.670	2.154	0.123
Residuals	72	1025.300	14.240		

Kruskal-Wallis Rank Sum Test			
	Test statistic	**Degrees of freedom**	**p-value**
Pollen	7.813	2	0.020
Nectar	21.699	2	<0.001

To identify the differences between crop types detected by the non-parametric analysis of variance Kriskal-Wallis test, we employ the non-parametric Nemenyi post-hoc test for multiple comparisons of rank sums. The table below; Table 23, gives p-values of the Nemenyi multiple comparisons tests of the RUD values between all pairs of crop types for pollen and nectar. The results suggest that statistically significant differences are detected between rapeseed and phacelia tanacetifolia (p-value<0.001) and other types of crop (p-value=0.001) for pollen RUD measurements, and between rapeseed and phacelia tanacetifolia (p-value=0.020) for nectar RUD measurements.

Table 23: Nemenyi's post hoc multiple comparisons test of rank sums in pollen and nectar between the types of crop.

Nemenyi's post hoc Multiple comparisons test (p-values)		
Pollen		
	Other	**Phacelia tanacetifolia**
Phacelia tanacetifolia	0.905	-
Rapeseed	0.001	<0.001
Nectar		
	Other	**Phacelia tanacetifolia**
Phacelia tanacetifolia	0.52	-
Rapeseed	0.55	0.020

The Nemenyi's post hoc multiple comparisons test results show that **differences are found in residue levels in nectar between *Rapeseed* and *Phacelia tanacetifolia*, and in pollen between *Rapeseed* and *Phacelia tanacetifolia*, and in pollen between *Rapeseed* and other types of crop.**

Tests of association

Table 24 shows tests of association between nectar and pollen for each type of crop. We note that strong positive correlation is detected between nectar and pollen for all three types of crop. The p-value in all Pearson and Spearman tests is significantly small and the null hypothesis of non-existence of correlation is rejected for all types of crop at significance level 5%.

Table 24: Correlation test between nectar and pollen for each type of crop.

Test for association/correlation between nectar and pollen					
	Pearson			Spearman	
	Correlation coefficient	95% Confidence Interval	p-value	Correlation coefficient	p-value
Nectar vs Pollen – Phacelia tanacetifolia	0.773	(0.571,1)	<0.001	0.386	0.038
Nectar vs Pollen - Rapeseed	0.802	(0.677,1)	<0.001	0.678	<0.001
Nectar vs Pollen - Other	0.984	(0.956,1)	<0.001	0.592	0.017

Location Comparison

In addition to tests of association, we also perform t-tests between the residue values in nectar and pollen for the three types of crop to determine whether the RUD values in nectar and pollen are different. The alternative hypothesis in the t-tests is that the mean RUD value in pollen is greater than in nectar. The results displayed on Table 25 suggest that both the Welch t-test and the non parametric Mann-Whitney test reject the null hypothesis for all types of crop.

Table 25: T-tests for mean comparison of RUD values in nectar and pollen for the different types of crop.

	F-test to compare variances		Welch two sample t-test				Mann-Whitney test
	Ratio of variances	p-value	t-statistic	Mean RUD pollen	Mean RUD nectar	p-value	p-value
Nectar vs Pollen – Phacelia Tanacetifolia	316.373	<0.001	2.942	43.975	2.064	0.007	<0.001

	F-test to compare variances		Welch two sample t-test				Mann-Whitney test
	Ratio of variances	p-value	t-statistic	Mean RUD pollen	Mean RUD nectar	p-value	p-value
Nectar vs Pollen - Rapeseed	724.280	<0.001	4.948	87.063	3.392	<0.001	<0.001
Nectar vs Pollen – Other	786.180	<0.001	2.795	50.153	1.051	0.014	<0.001

Regression analysis

As a next step we perform linear regression in order to model the relationship between the RUD values in nectar and pollen for the different types of crop. The results of the regression analysis are presented in Table 26 below. Recall that the model examined is of the form

$$Nectar_{RUD} = a + bPollen_{RUD}$$

where a is the intercept and b is the change in the RUD in nectar caused by a unit change in the RUD in pollen.

Table 26: Linear regression between RUD values in nectar and pollen for the different types of crop.

	Coefficients		Std. Error	t-value	p-value	R^2	Residual Std. Error
Nectar vs Pollen – Phacelia tanacetifolia	$\hat{a}$	0.250	0.725	0.345	0.733	0.597	2.907
	$\hat{b}$	0.041	0.008	5.440	<0.001		
Nectar vs Pollen - Rapeseed	$\hat{a}$	0.796	0.501	1.59	0.121	0.644	2.312
	$\hat{b}$	0.030	0.004	7.952	<0.001		
Nectar vs Pollen - Other	$\hat{a}$	-0.880	0.162	-5.428	<0.001	0.969	0.447
	$\hat{b}$	0.034	0.002	18.498	<0.001		

In addition to the results of the previous analyses per crop type, the regression analysis further supports the findings. Accordingly, the estimated regression lines are given in Table 27 and displayed in Figure 20.

Table 27: Estimated regression lines per type of crop.

	Estimated regression line
Nectar vs Pollen – Phacelia tanacetifolia	$\widehat{Nectar}_{RUD} = 0.250 + 0.041 Pollen_{RUD}$
Nectar vs flower - Rapeseed	$\widehat{Nectar}_{RUD} = 0.796 + 0.030 Pollen_{RUD}$
Nectar vs Pollen - Other	$\widehat{Nectar}_{RUD} = -0.880 + 0.034 Pollen_{RUD}$

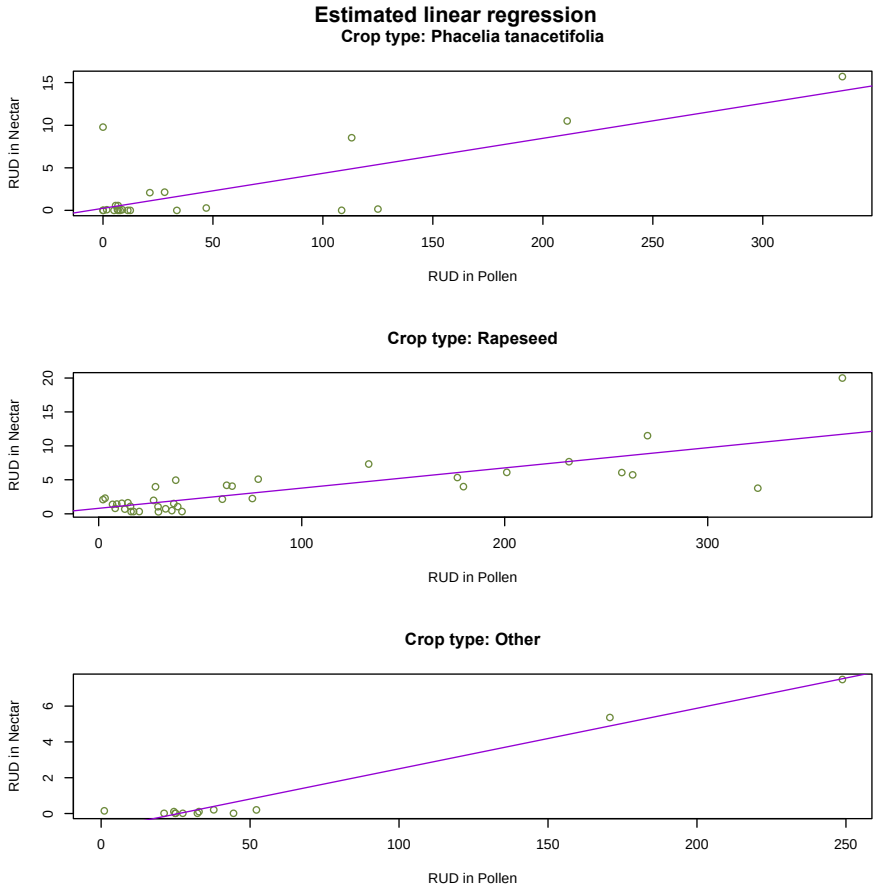


Figure 20: Estimated linear regression of nectar-pollen for the different types of crop.

In conclusion:

- The residue levels in rapeseed notably exceed the residue levels in *Phacelia tanacetifolia* and other types of crop, both for pollen and nectar.
- For all types of crop, the residue in pollen is higher than the residue in nectar, irrespective of the sampling matrix.
- Positive correlation between nectar and pollen exists, stronger in the case of rapeseed.
- Statistically significant differences in the mean residue values in nectar and pollen are detected for all types of crop, with the difference being greater in the case of rapeseed.

4.1.4. RUD in pollen and nectar – Direction of pesticide application related differences

The number of available observations does not allow investigation of direction of pesticide application related differences, as there exist 81 observations in nectar and pollen in which the direction of the application of the pesticide is *downward* and only 3 observations with *sideward* direction of application.

4.1.5. RUD in pollen and nectar – Substance related differences

The prime focus of this section is to examine the existence of substance related differences in the RUD values in nectar and pollen. The aim is to identify which substances affect the residue levels in nectar and pollen.

Table 28 presents a summary of the residue values in nectar and pollen for the active substances for which more than 5 observations are available from the data.

Not enough observations were available for:

- **Pollen**: for the substances 2,4 D, Folpet, Fluopyram, Flupyradifurone, Trifloxystrobin, Chlorantraniliprole, Glyphosate, Spirotetramat, Thiram (1 observation), Beta-cyfluthrin, Iprodione (2 observations), Gamma-cyhalothrin, Indoxacarb (3 observations)
- **Nectar**: for the substances 2,4 D, Folpet, Fluopyram, Flupyradifurone, Trifloxystrobin. Chlorantraniliprole, Glyphosate (1 observation), Beta-cyfluthrin, Iprodione (2 observations), Cyantraniliprole, Gamma-cyhalothrin, Indoxacarb (3 observations)

Table 28: Summary statistics of the residue values in nectar and pollen for the active substances for which more than 5 observations are available

Substance	Min.	1st Quartile	Median	Mean	3rd Quartile	Max.	St. Dev.	# of obs.
Pollen								
Cyclaniliprole	0.003	3.788	7.775	20.620	23.000	108.500	31.847	11
Dimoxystrobin	24.440	33.640	37.380	40.060	38.670	78.560	15.590	10
Boscalid	21.130	27.560	29.300	33.540	32.790	65.710	12.866	10
Metconazole	6.790	8.090	8.897	22.770	27.030	63.030	23.990	5
Pyraclostrobine	12.830	14.340	15.610	35.870	60.880	75.680	30.061	5
Alpha-cypermethrin	11.370	30.480	179.700	167.300	260.300	366.300	121.438	15
Cyantraniliprole	1.071	9.158	13.720	17.120	28.740	32.820	13.131	6
Nectar								
Cyclaniliprole	0.0001	0.0001	0.001	0.013	0.003	0.125	0.039	10
Dimoxystrobin	0.010	0.128	0.339	1.370	1.394	5.100	1.983	10
Boscalid	0.010	0.010	0.146	1.015	0.961	4.071	1.626	10
Metconazole	0.795	1.383	1.423	1.955	1.979	4.195	1.321	5
Pyraclostrobine	0.680	1.092	1.626	1.560	2.156	2.245	0.675	5
Alpha-cypermethrin	0.333	0.938	5.333	5.358	6.717	20.000	5.227	15

We result that the substance that leaves the highest residue in both pollen and nectar is Alpha-cypermethrin. Figure 21 displays boxplots of the residue values in pollen and nectar for the

substances for which more than 5 observations are available. We see that Alpha-cypermethrin is the substance with the highest residue values in both pollen and nectar, while Pyraclostrobine and Metconazole in the case of nectar, and Dimoxystrobin, Boscalid in the case of pollen indicate high residue values.

We note however, that the substances 2,4 D, Fluopyram, Flupyradifurone, Trifloxystrobin, Glyphosate (nectar and pollen) and Thiram (only for pollen), for which only one residue observation is available, require further data collection and examination, considering that the observation in the dataset is remarkably high, and in fact higher than the residue values observed for Alpha-cypermethrin.

Furthermore, we do not examine in this section substance related differences for each of the five sampling matrices due to the lack of a number of observations for each sampling matrix adequate to result to reliable statistics.

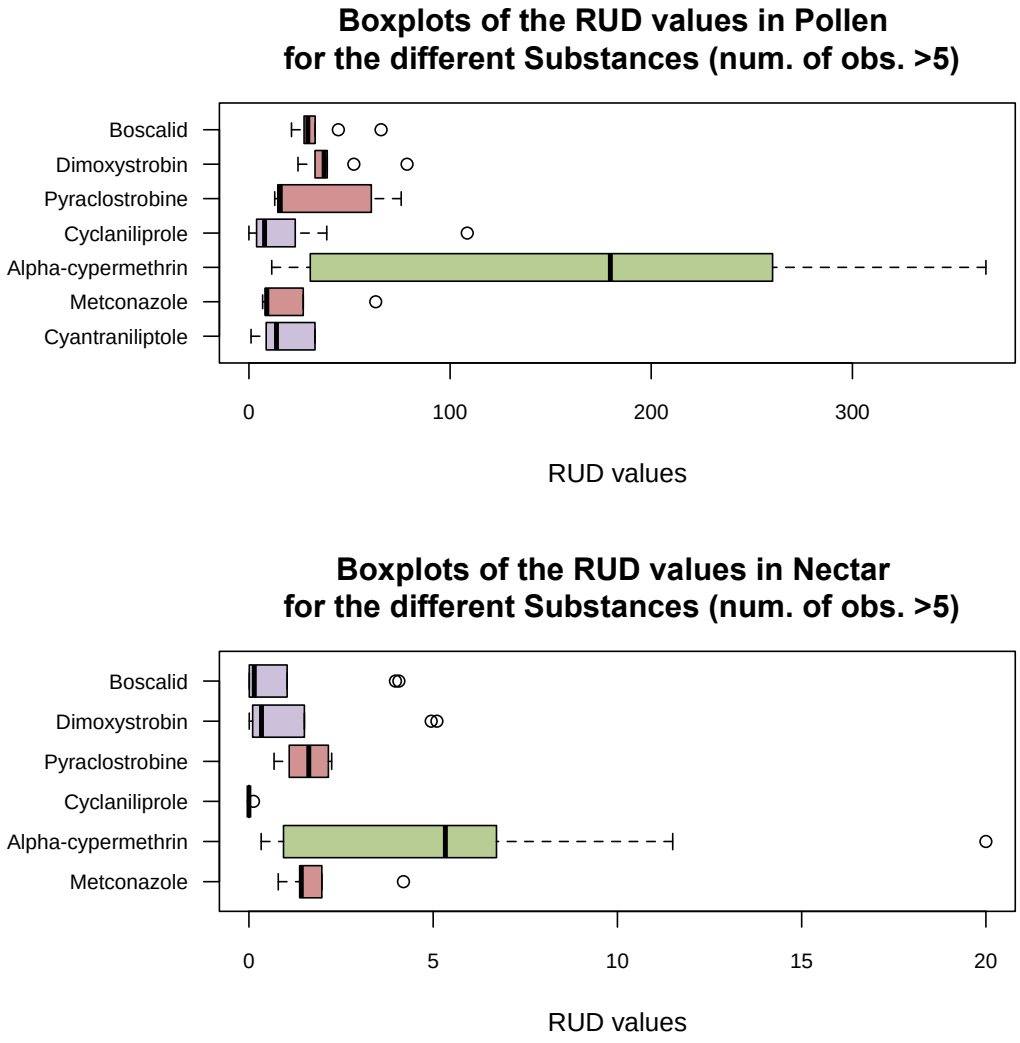


Figure 21: Boxplots of the RUD values in Pollen and Nectar for the different substances. Only substances for which more than 5 observations are available are considered.

Analysis of variance

In addition to descriptive analysis we consider at this point analysis of variance in the residue values in nectar and pollen to test whether there exist statistically significant differences among substances. The results of the ANOVA test are presented in Table 29 below.

Table 29: Analysis of variance (ANOVA) table of the residue values on the substance.

Analysis of variance (ANOVA) table

	Degrees of freedom	Sum of Squares	Mean Sum of Squares	F-statistic	p-value
Pollen					
Type of crop	18	403273	22404	5.983	<0.001
Residuals	61	228422	3745		
Nectar					
Type of crop	18	591	32.830	3.709	<0.001
Residuals	56	495.700	8.850		

Kruskal-Wallis Rank Sum Test

	Test statistic	Degrees of freedom	p-value
Pollen	44.935	18	<0.001
Nectar	49.365	18	<0.001

The analysis of variance (ANOVA and non-parametric Kruskal-Wallis test) detects a statistically significant difference in the residue values among substances. However, the available observations are few and post hoc tests to identify which substances differ, would not result to statistically reliable outcomes.

Concluding, we are significantly restricted in performing other tests to compare the residue in nectar and pollen for the various substances, due to the very small sample sizes for each substance and the violation of the normality assumption.

In conclusion:

- Statistically significant substance related differences are detected in the residue levels in nectar and pollen.
- We examine substances with more than 5 observations available. The substance that appears to leave the highest residue in both nectar and pollen is Alpha-cypermethrin. However, there exist substances with higher residue values but fewer observations.
- The number of observations available for each substance is not enough to produce reliable statistics and further investigation is required.

4.2. RUD levels in pollen and nectar in comparison with physicochemical and environmental properties of the active substance

In this section we focus on the third objective of the data analysis, that is on investigation of potential correlations between residue levels in nectar or pollen with the physicochemical and environmental properties of the active substance. The substances included in the analysis are Alpha-cypermethrin, Beta-cythrucin, Boscalid, 2,4 D, Flupyradifurone, Indoxacard, Cyantraniliprole, Folpet, Metconazole, Cyclaniliprole, Gamma-cyhalothrin, Pyraclostrobine, Thiram, Chlorntraniliprole, Iprodione, Glyphosate and Dimoxystrobin.

In particular, we consider the solubility in water in (mg/L) and the octanol water partition coefficient (logPow); for both properties values at pH 7 were used for data analysis. In case no value is available at pH 7, the available value (at any pH) was used. Arithmetic mean value of Soil Organic Carbon-Water Partitioning Coefficient (Koc), if feasible to be derived, was considered as well. Moreover, the different methods of application are considered separately. To investigate the association between residue levels in nectar or pollen with the physicochemical properties of the active substance, the RUD values in nectar or pollen have been paired with the corresponding solubility, log Pow and Koc values of the active substance used. The limited number of observations for each active substance made it statistically quite difficult to further investigate potential correlations per active substance.

Two large (but true) solubility values are observed. The first corresponds to glyphosate used in study 2311016/V7YH1002 where the solubility of glyphosate in water is 10.5g/L, and the second one corresponds to the active substance 2,4 D used in study S11-02084, for which the solubility value is 24.3g/L.

As already mentioned, the direction of application methods are considered separately for the correlation analysis. Two application methods, downward and sideward, are included in the dataset, however only three observations are available for which sideward application was employed. As a consequence, only for downward application correlation analysis can be performed.

In the following, we display scatterplots of the RUD values in nectar and pollen to the physicochemical characteristics of the active substances; see Figure 22, as well as the results of the correlation analysis of the residue in nectar and pollen with the physicochemical and environmental properties; see Table 30.

The plots in the top row of Figure 22 display the values of the residue levels in nectar and pollen to the corresponding values of solubility in water. Because of the large solubility values present in the dataset, the two right plots provide a closer look to the remaining data points. The bottom plots of Figure 22 correspond to the RUD in nectar and pollen to the logPow and Koc values.

The results of the correlation analysis displayed in Table 30 show that no correlation can be identified between the residue in pollen and solubility in water of the active substance, since both the Pearson and Spearman correlation tests cannot reject the null hypothesis of no correlation. On the contrary, the tests reject the null hypothesis in the case of nectar and indicate the presence of weak positive correlation between the residue levels in nectar and solubility in water of the active substance.

Both correlation tests fail to reject the null hypothesis of no correlation (p-value>0.005) in the comparison between the residue in pollen and the logPow values and so the two are uncorrelated. On the other hand, the null hypothesis is rejected in the case of the RUD in nectar and Koc values, indicating weak positive correlation between the two.

Concrete and reliable conclusions however cannot be derived in terms of the relationship between the residue in nectar and logPow values, and residue in Pollen with Koc values. That is reflected through the two different correlation coefficients that try to capture their relationship but results in contradictory conclusions.

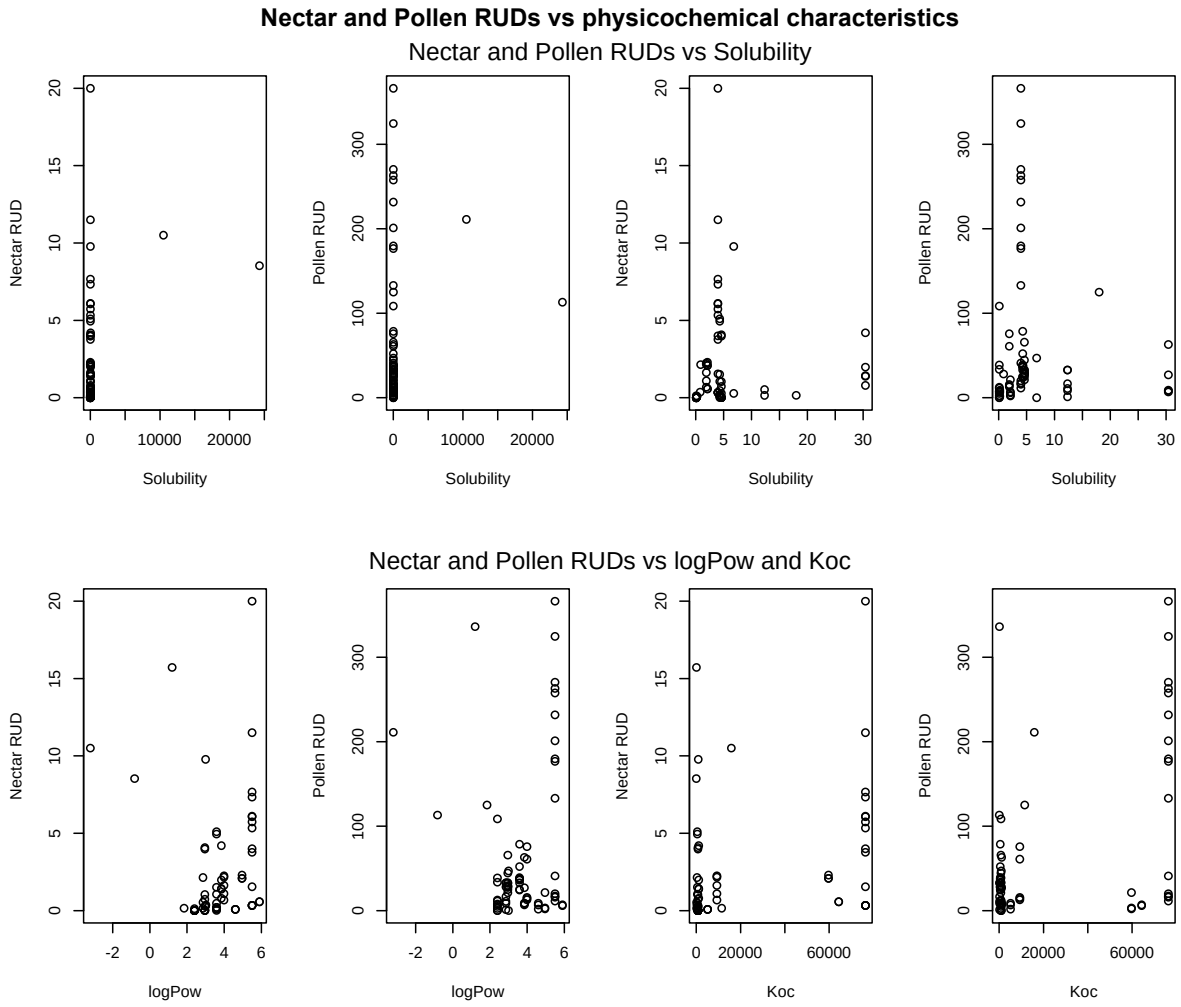


Figure 22: Boxplots of the RUD values in Pollen and Nectar for the different substances. Only substances for which more than 5 observations are available are considered.

Table 30: Correlation test between nectar and pollen and the physicochemical and environmental properties of the active substance, with downward direction of application of the pesticide.

Test for association/correlation					
	Pearson			Spearman	
	Correlation coefficient	95% Confidence Interval	p-value	Correlation coefficient	p-value
Nectar vs Solubility	0.309	(0.084,0.505)	0.008	0.321	0.006
Pollen vs Solubility	0.157	(-0.071,0.369)	0.175	0.218	0.068
Nectar vs log Pow	-0.014	(-0.243,0.217)	0.908	0.435	<0.001

Test for association/correlation					
Pollen vs log Pow	0.125	(-0.102,0.339)	0.280	0.135	0.243
Nectar vs Koc	0.346	(0.236,0.534)	0.003	0.323	0.005
Pollen vs Koc	0.496	(0.306,0.648)	<0.001	0.114	0.323

In conclusion:

- Weak positive correlation is detected between the residue levels in nectar and solubility in water, and between the residue levels in nectar and Koc values of the active substance.
- No correlation can be detected between the residue levels in pollen and solubility in water, and between the residue levels in pollen and log Pow.
- Concrete results cannot be derived in the cases of residue in nectar and log Pow values, and residue in pollen and Koc values.

4.3. Residue decline analysis in pollen and nectar in comparison with residue decline in other environmental matrices

This section focuses on the fourth objective of the data analysis, namely on the investigation of potential correlations between residue decline (DT_{50}) in pollen and nectar and other environmental matrices, namely residue decline in soil and water.

To investigate the association between the residue decline in nectar or pollen and the residue decline in soil and water, the DT_{50} values in nectar or pollen have been paired with the corresponding DT_{50} values in soil and water of the active substance used. The limited number of observations for each active substance made it statistically quite difficult to further investigate potential correlations per active substance.

In the following, we present correlation tests results between the DT_{50} in nectar and pollen, and the DT_{50} in soil and water; see Table 31. The DT_{50} values are expressed in days. Considering that we have available 8 observations for residue decline in nectar and 19 observations for residue decline in pollen, we conduct both the Pearson and the Spearman correlation tests. The results presented in the table suggest that the null hypothesis of the true correlation being zero cannot be rejected. Therefore we conclude that the residue decline in nectar and pollen is uncorrelated with the residue decline in soil and in water.

Table 31: Correlation test between DT_{50} values in nectar and pollen, soil and water.

Test for association/correlation					
	Pearson			**Spearman**	
	Correlation coefficient	**95% Confidence Interval**	**p-value**	**Correlation coefficient**	**p-value**
DT_{50} in nectar vs soil	0.493	(-0.325,0.889)	0.215	0.450	0.264
DT_{50} in pollen vs soil	0.046	(-0.417,0.489)	0.853	0.091	0.713

Test for association/correlation					
DT_{50} in nectar vs water	0.428	(-0.396,0.870)	0.290	0.517	0.189
DT_{50} in pollen vs water	-0.108	(-0.536,0.364)	0.659	-0.202	0.407
DT_{50} all observations vs soil	0.0859	(-0.304,0.451)	0.670	0.209	0.296
DT_{50} all observations vs water	-0.035	(-0.409,0.349)	0.862	-0.077	0.702

In conclusion:

- The residue decline, calculated by the DT_{50} in days, in nectar and pollen is uncorrelated with the residue decline in soil and water.

5. Overall conclusions

In the frames of the project "Collection and analysis of pesticide residue data for pollen and nectar", carried out under the contract OC/EFSA/PRAS/2015/08, new available information derived from residue trials were collected, evaluated, used in order to create a database for pesticide residue levels and residue decline in pollen and nectar and finally considered to perform a data analysis with relevant parameters.

Firstly, a detailed screening of regulatory documents was carried out in order to identify appropriate studies with residue analysis in pollen, nectar and other relevant matrices as well as estimation of sugar content of nectar and protein content of pollen. During the screening procedure 314 pesticide active substances were screened (AIR II, AIR III and New Active Substances) and 125 relevant studies have been identified.

The number of identified studies per active substance is presented in Table 32 and Figure 23 below:

Table 32: Number of identified studies included in the database per active substance

Active substance	Number of selected/relevant studies per active substance
2,4 D	1
Acetamiprid	5
Alpha-cypermethrin	2
Beta- cyfluthrin	2
Boscalid	2
Chlorantraniliprole	3
Clothianidin	24
Cyantraniliprole	29
Cyclaniliprole	6
Dimoxystrobin	2
Emamectin	2
Ethephon	1
Fluopyram	2
Flupyradifurone	13
Folpet	1
Gamma-cyhalothrin	4

Active substance	Number of selected/relevant studies per active substance
Glyphosate	1
Indoxacarb	7
Iprodione	2
Metconazole	1
Oxamyl	1
Pymetrozine	9
Pyraclostrobin	1
Spirotetramat	2
Tau-fluvalinate	1
Tebuconazole	1
Thiacloprid	1
Thiamethoxam	1
Thiram	1
Trifloxystrobin	2
TOTAL	**130***

* 5 studies have been conducted with test items containing 2 active substances

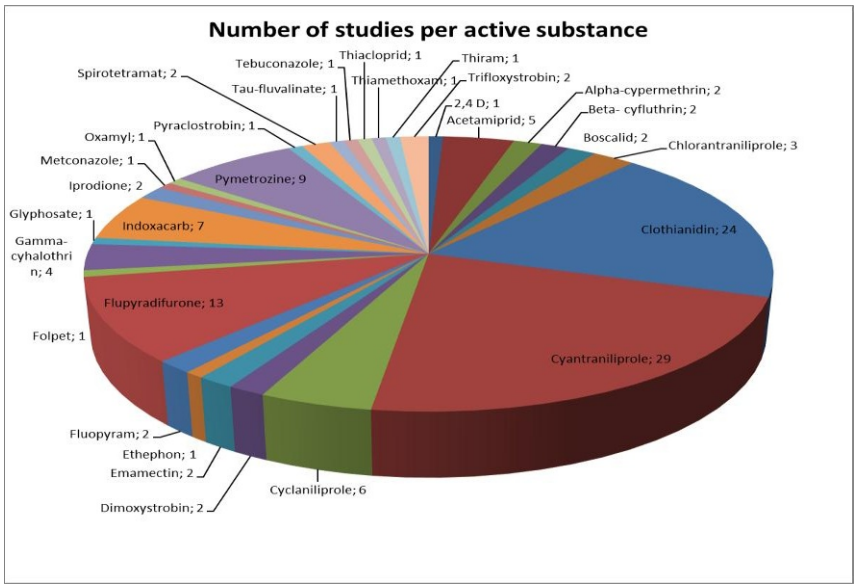


Figure 23: Number of relevant studies in the database per active substance.

As regards the category of these active substances, the vast majority of studies were carried out with insecticides (Figure 24) as it was expected, since these studies are used as field studies usually needed for insecticides higher tier risk assessment.

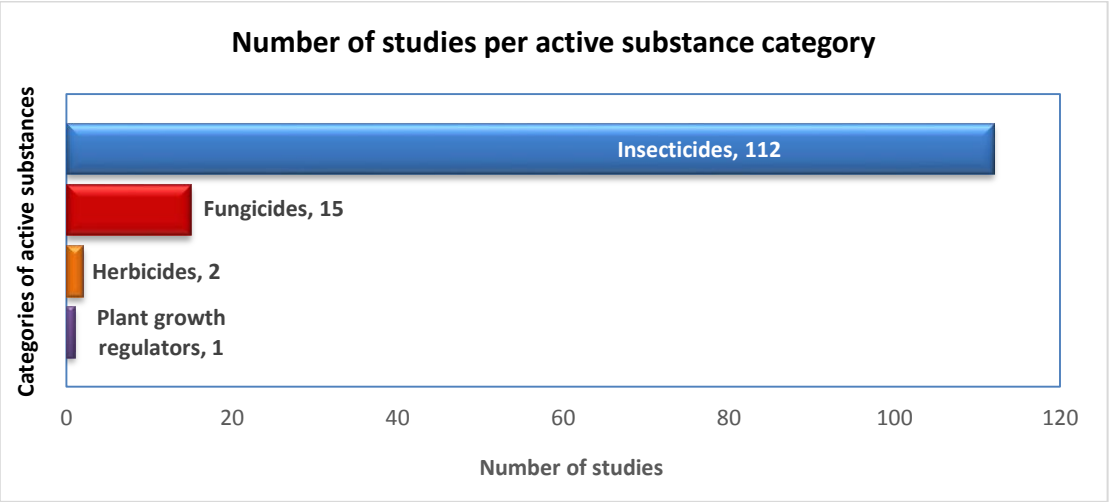


Figure 24: Number of studies per active substance category.

Subsequently, each identified study was evaluated according to the assessment protocol developed in the frame of this project [based on the principles of Appendices G and S of the EFSA GD (EFSA Journal 2013:11(7): 3295)] and allocated in one of the four predefined categories as described in section 3.4. It is noted that the evaluation was focused on parameters and elements relevant for this specific project and the final conclusion regarding the reliability of each study is applicable only to the use of the study in the frames of the project. This means that even if a study has been characterized as "not reliable" within this project, it will not have any influence on the use of this study during the peer review process of an active substance under Reg (EC) 1107/2009.

In several studies many deficiencies were identified and the most important and frequently observed were the following:

- Inadequate reporting of the study design (trial conditions, crop related parameters, application parameters, sampling methodology, *etc*)
- Fewer than recommended sampling time points (in some cases only one or two samplings after application were carried out)
- First sampling not immediately after the application, but a few days after the last application
- More than one application in the same trial/plot
- More than one type of application in the same trial/plot (e.g. spray foliar application and seed treatment, granule application on bare soil and spray foliar application)
- Inadequate data regarding the spatial distribution of samples collected directly from the crop
- Not appropriate analysed matrices, in order to be able to calculate the Landscape Dilution Factor (i.e. pollen and/or nectar from bees and pollen and/or nectar from crop).

These limitations resulting in inability to:

- derive reliable and worst case RUD values to be used for risk assessment
- calculate reliable DT_{50} values for pollen, nectar and relevant matrices
- calculate the Landscape Dilution Factor (LDF)

Nevertheless, there were many studies that fulfilled the predefined criteria and RUD and DT_{50} values were calculated as presented in more details in previous sections of this report.

The RUD values derived by reliable studies were further evaluated according to criteria discussed and agreed with EFSA and only some of these values were considered reliable and appropriate in order to be further used for (i) data analysis performed in the frame of this project and (ii) enrichment of the data set of RUD values already available in Appendix F of the EFSA bees GD [EFSA Journal 2013:11(7): 3295] and used for pollinators risk assessment.

The most robust data set was derived by studies with spray foliar application of pesticides. Several reliable RUD values were calculated for nectar and pollen for several active substances. For all relevant matrices (nectar and pollen collected from flowers or/and from bees/traps) the highest RUD values were observed immediately after the application on 0DAA or 1DAA. High RUDs were calculated in the majority of the active substances, while the initial worst case RUDs for other substances were significantly lower. It is acknowledged that these differences maybe occur due to different crops, different time between the application and the first sampling or other parameters used in each study. In all cases a fast decline in the residues was observed and the RUD values calculated from samples taken later than the 2nd day after application were much lower than the initial values.

For granule applications the only reliable RUD values were derived from studies with the active substance clothianidin. The RUDs in pollen collected from different sources (i.e. bees, pollen traps and flowers) were comparable and no decline was observed in samples taken in different time points after the start of flowering.

For seed treatment not reliable RUD values were derived for pollen or nectar, since the only available residue data were collected from not reliable studies (studies with only one sampling time point or studies with inadequate reporting of the study design in the study report).

In addition, reliable DT_{50} values were calculated for pollen and nectar for several active substances. The calculated DT_{50} values were in most of the cases less than 2 days for both matrices (pollen and nectar). In some cases where an FOMC kinetic fit was considered, DT_{50} values in pollen were estimated to be approximately 4 days. Nevertheless, these values are much lower than the default value of 10 days that is used according to the EFSA GD [EFSA Journal 2013:11(7): 3295].

Furthermore, in the frame of this project a data analysis was performed covering four main objectives in order to investigate potential correlations between:

(i) residue levels (RUDs) in pollen or nectar versus residue levels in plant foliage

(ii) residue levels (RUDs) in pollen versus residue levels in nectar;

(iii) residue levels (RUDs) in pollen or nectar versus physical-chemical properties of the active substance (S, logPow, Koc);

(iv) residue decline in pollen or nectar versus residue decline in different environmental compartments (i.e. soil, water)

As regards residue levels comparison, statistically significant differences were detected between nectar, pollen and plant foliage. Particularly, the residue levels in nectar were lower than both pollen and plant foliage, while the highest residue levels were observed in pollen.

Comparing the residue levels in nectar and pollen, we observed that the residue levels in pollen were higher than the residue levels in nectar, irrespective of both the sampling matrix (flower/bees/traps) and the type of crop (rapeseed/*phacelia tanacetifolia*/other). Statistically significant differences were detected among sampling matrices, with the residue levels in both pollen and nectar being highest when extracted from flower. Further, strong positive correlation was observed between nectar and

pollen for all types of sampling matrices, with the largest correlation being detected when nectar and pollen are extracted directly form the bees. As regards the type of crop, the residue levels in rapeseed notably exceeded the residue levels in *Phacelia tanacetifolia* and other types of crop, both for pollen and nectar. Furthermore, positive correlation was found between nectar and pollen which is stronger when the type of crop was rapeseed.

Statistically significant differences have been identified between plant foliage and pollen, and plant foliage and nectar. More specifically, the mean residue in pollen was higher than the mean residue in plant foliage, and the mean residue in plant foliage was higher than the mean residue in nectar. On the other hand, there did not exist statistically significant differences between pollen from bees with plant foliage. Unfortunately, the possible effect of the direction (downward vs sideward application) of the pesticide application on RUD levels in nectar and pollen was not possible to be studied, because the number of available observations of sideward application was significantly small; only 3 out of 84 observations.

Substance related differences have been detected in the residue levels in nectar and pollen. In all cases the number of available observations for each substance was notably small and produced results should be interpreted with caution. To be more precise, apha-cypermethrin was the substance that appears to leave the highest residue in both pollen and nectar; however, the substances 2,4 D, fluopyram, flupyradifurom, trifloxystrobin and glyphosate, for which only one observation was available, require further data collection considering that the corresponding observation in the dataset was remarkably high.

Regarding investigation of possible correlations between residue levels in nectar and pollen, and physicochemical and environmental properties of the active substance the main focus was on the solubility in water, logPow and Koc value in water. The results revealed a weak positive correlation between the residue levels in nectar with solubility and Koc values. No correlation has been detected between the residue levels in pollen with solubility in water and logPow values. However, no concrete results could be identified for the residue in nectar and logPow, and the residue in pollen and Koc values.

Finally, possible correlations between residue decline (DT_{50} values) in nectar or pollen with residue decline in soil and water have been examined. The analysis showed that the residue decline values between nectar and pollen with soil and water were uncorrelated.

In conclusion, in the frames of the project "Collection and analysis of pesticide residue data for pollen and nectar", carried out under the contract OC/EFSA/PRAS/2015/08, new available residue data for pollen, nectar and related matrices were collected in a database especially developed for this purpose. From all collected data, several reliable RUD and DT_{50} values in relevant matrices were calculated and can be used to address identified gaps and to update the data set used for risk assessment for pollinators according to the EFSA GD [EFSA Journal 2013:11(7): 3295] in order to enhance the 1st tier assessment reliability.

References

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European Food Safety Authority, 2013. Guidance on the risk assessment of plant protection products on bees (Apis mellifera, Bombus spp. and solitary bees). EFSA Journal 2013;11(7):3295, 266 pp. doi:10.2903/j.efsa.2013.3295.

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Abbreviations

a.s.	active substance
BPI	Benaki Phytopathological Institute
CS	Capsule suspensions
DAR	Draft Assessment Report
DB	Database
DFOP	Double First-Order in Parallel
DT_{50}	Degradation/Dissipation Time (for 50% disappearance)
DT_{90}	Degradation/Dissipation Time (for 90% disappearance)
EC	European Commission
EC	Emulsifiable concentrate
EFSA	European Food Safety Authority
EU	European Union
FOMC	First Order Multi-Compartment
GD	Guidance Document
GLP	Good Laboratory Practice
ID	Identification / Identity
IQR	Interquartile range
Koc	Organic carbon – water equilibrium partition coefficient
LOD	Limit of detection
LOQ	Limit of quantification
ME	Microemulsion
MRL	Maximum Residue Limits
OD	Oil dispersion
PHI	Post Harvest Interval
Pow	Octanol/water partition coefficient
PPPs	Plant Protection Products
RAR	Renewal Assessment Report
RR	Registration Report
RUDs	Residue Unit Doses
SC	Suspension concentrate
SE	Suspo-emulsion
SFO	Single First Order
SG	Water soluble granule

SL	Soluble (liquid) concentrate
US EPA	United States Environmental Protection Agency
WG	Water dispersible granules
WP	Work Package
WP	Wettable powder

Appendix A – Assessment protocol

Assessment protocol for the evaluation of the identified studies.

For the file, see attachments to this report.

Appendix B – List of active substances and relevant documents screened

List of active substances and relevant regulatory documents screened (in Excel format)

For the file, see attachments to this report.

Appendix C – List of identified relevant studies

List of identified relevant studies (in Excel format)

For the file, see attachments to this report.

Appendix D – List of identified relevant studies in an ENDNOTE library file

List of identified relevant studies (in ENDNOTE format)

For the file, see attachments to this report.

Appendix E – Evaluation check lists

Evaluation check listed for the selected studies

For the file, see attachments to this report.

Appendix F – Completed database

OC/EFSA/PRAS/2015/08 Excel Database Pollen & Nectar

For the file, see attachments to this report.

Appendix G – Brief description of the selected studies

Brief description summarizing the main points of each one of the selected studies

For the file, see attachments to this report.

Appendix H – Raw data for DT_{50} and DT_{90} calculations

Raw data for DT_{50} and DT_{90} calculations

For the file, see attachments to this report.