



EPQ – Requirement No. 5

Deliverable D10.5

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1 WR, 2 UGENT

B-GOOD

Giving Beekeeping Guidance by cOmputatiOnal-assisted Decision making



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Preface

This deliverable is one out of five related to ethics requirements. It addresses ethics issues with respect to environmental protection and safety within B-GOOD.

Summary

This deliverable provides further information about the possible harm to the environment that may be caused by the research activities within B-GOOD and about the measures that will be taken to mitigate the risks. These risks relate to the use of insecticides for research purposes. Health and safety procedures conforming to relevant local/national guidelines/legislation are described. Confirmation is provided that copies of authorisations for relevant facilities are kept on file.

1. Assessment of ethics issues

1.1 Background

B-GOOD's key focus is on healthy and sustainable beekeeping. The B-GOOD project involves studies with honey bees (*Apis mellifera*). Some of the honey bee colonies within B-GOOD will be exposed to induced stressors (insecticide exposure, low food availability, *Varroa destructor* parasitic mite infestation, queen failure) in such a way that effects on their health are to be expected. These experiments, including insecticide exposure studies are essential to validate the tools that are developed within B-GOOD.

However, insecticide exposure of honeybee colonies may cause risks to the environment around the exposed colonies and to the people applying it. Therefore, the research involving insecticides entails possible risks related to environmental protection and safety that are not covered by standard beekeeping protocols or interventions.

The concerned studies are part of B-GOOD work package 1. The stress-exposed colonies will be restricted to Tier 1 within Pilot B, and will be performed at one location by one partner (partner WR, country: the Netherlands), which has ample experience in conducting such work (e.g. Blanken et al., 2015, Van Dooremalen et al., 2018). Pilot B will last for three years, from month 7 to month 42. The insecticide exposure will be addressed as described below.

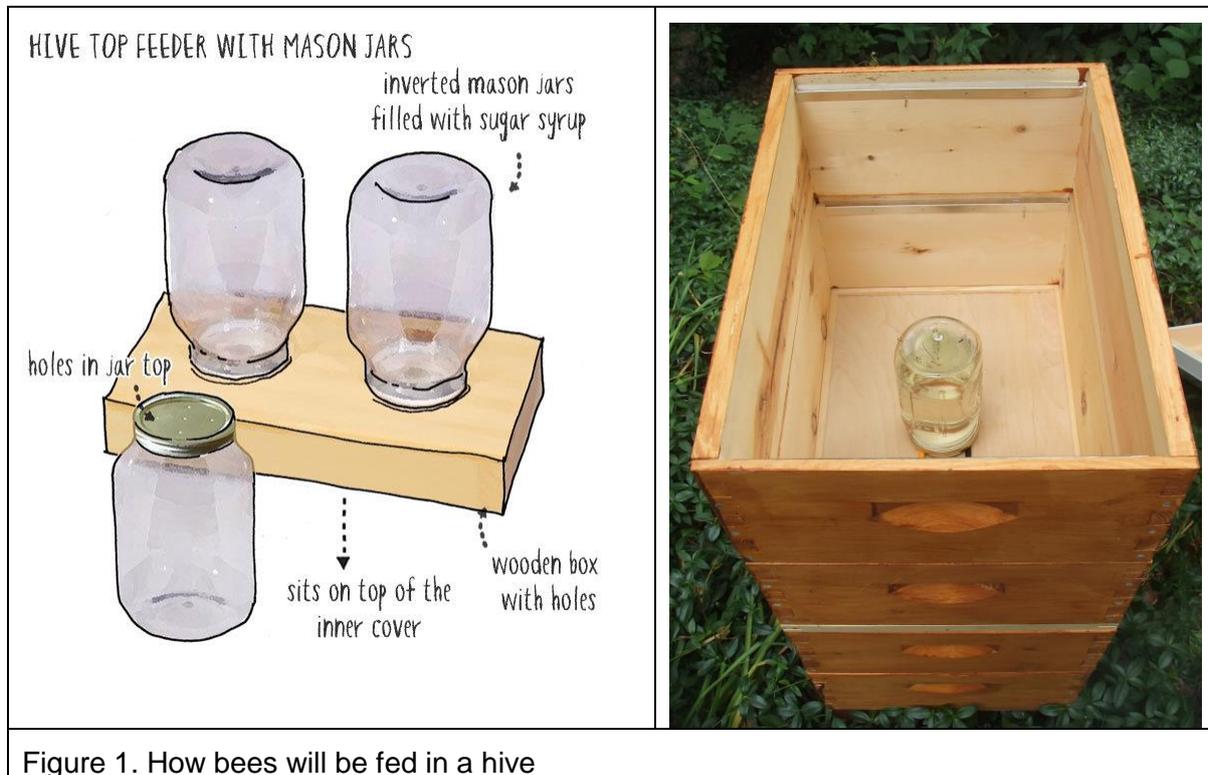
1.2 Possible harm to the environment and risk mitigation

Within B-GOOD, five honey bee (*Apis mellifera*) colonies will be persistently exposed to sub-lethal doses of a number of insecticides in order to validate whether B-GOOD's new tools to monitor bee health pick up a reduced colony health status as a result of the pesticide application (WP1, task 1.2, pilot study B). Insecticides will be selected to reflect future risks for bee losses. At the moment of writing, the concept list of insecticides consists of Acetamiprid, Sulfoxaflor, and Pyrethroids.

All insecticide exposures will be performed at the facilities of partner [WR in the Netherlands](#), at Wageningen Plant Research. WR is an accredited research institute permitted to use insecticides for research purposes. At the institutional level, two people are appointed at Wageningen Plant Research as biological safety experts (Reinoud Bouwer and Carolien Ruyter). Exposure will be undertaken in accordance with relevant regulations and legislation (e.g. related to the removal of waste materials), and taking into account appropriate protection of research staff and the environment.

Specifically, five colonies will be exposed between April and November (2020-2022) to acute and chronic doses of insecticides throughout the honey bee flight season (this is, when bee resources are available and bees go out to forage). Risk of environmental contamination is estimated to be very low as the following risk mitigation measures will be taken:

- The pure compounds are directly applied to bee colonies for research purpose, and not in the open. These colonies are kept outside in an appointed apiary without access by third parties.
- Exposure will occur via sugar water that will be offered to the colonies directly inside each hive (Figure 1). This exposure route will ensure that only the honey bees of that specific hive will have access to this source.



- Stock solutions and feeding solutions are prepared at the laboratory, in a bee free room, and moved to the apiary before feeding. While outside, containers are covered. These measures are taken to prevent exposure to robbing bees.
- Spare containers that are not used and containers with leftovers from the previous feeding occurrence are collected and returned to the laboratory.
- Waste disposal will be done according to standard procedures, i.e. collecting contaminated solutions. Rules applicable to plant protection products, skin invaders and carcinogens, all fall under the "Extremely toxic 05" waste stream with a separate and dedicated-for-this-purpose 'separated liquid and solid waste stream' and its own mandatory prescribed packaging (see Appendix 1).
- One potential route of insecticides entering the environment is via bees that feed on the sugar solution inside the hive, go outside the hive to forage and die there. This cannot be prevented. Concentrations traced in bees from exposed colonies are much lower than the fed concentrations (Van Dooremalen et al., 2018), therefore the expected environmental contamination will be very low. To mitigate environmental contamination via this route, dead bees that may be found directly in front of exposed hives, will be removed to stop other organisms (e.g. birds, mice, wasps, where wasps are the organisms most at risk) from feeding on them.

1.3 Health and safety procedures for staff involved

The beneficiary confirms that appropriate health and safety procedures conforming to local/national guidelines/legislation will be followed for staff involved in the research involving insecticide exposure of bee colonies (WR institute).

Mandatory rules apply to all employees and guests with regard to safety and laboratory, field and greenhouse work. Related documents can be found digitally by employees on institutional internal share folders and via posters at the dedicated working areas. At the institutional level, one person is appointed as safety expert (Arjo Meijering) for questions and support of the organisation's first aid team in case of incidents at Wageningen Plant Research. Additional guidelines include:

- Safe and general work rules related to the Wageningen Research buildings (drawn up in 2015) and its laboratories (drawn up 25 April 2019) with corresponding mandatory lab tests for new permanent and temporary employees.
- Various safety manuals and protocols about biosafety, general lab safety, equipment and machines, chemicals, waste management and mandatory clothing regulations.
- The 10 main rules of the Practical Guide to the Lab (Puylaert et al 2011) apply also for working with insecticides.
- A mandatory Bee Training protocol regarding bee stings and how to act (Appendix 2) applies specifically for bee work.
- As a general guideline, no single-person bee handling is permitted. Hence, working with the bees will always be done by a team of minimum two persons.

For working safely with chemicals, reference is made to the

- labeling on the chemical packaging;
- package leaflet (MSDS) from the supplier;
- Internet; Gros system;
- ChemWatch software. WR has a subscription for Chemcards.

In addition to standard rules, protocols and regulations, specifically for the exposed colonies: honey harvested (if needed) will be collected separate from other colonies for research purpose only, be labelled as hazardous waste and be disposed of similarly to feeding solutions (see above). This honey will NOT be used for human consumption.

2. Confirmations

The B-GOOD consortium confirms that copies of authorisations for relevant facilities – in this case, biosecurity classifications of laboratories involved in the research activities, and as far as applicable – are kept on file and will be made available upon request.

3. References

Blanken, L. J., van Langevelde, F., & van Dooremalen, C. (2015). Interaction between *Varroa destructor* and imidacloprid reduces flight capacity of honeybees. *Proceedings of the Royal Society B: Biological Sciences*, 282(1820), 20151738.

Puylaert, P.G.B., Verwaaijen, B., Brouns, S.J.J., Swarts, D.C., Bogert, T. van den, Ryback, B. (2015). A practical guide to the lab. Second edition (version 2.1), 126p. Wageningen University

van Dooremalen, C., Cornelissen, B., Poleij-Hok-Ahin, C., & Blacquièrè, T. (2018). Single and interactive effects of *Varroa destructor*, *Nosema* spp., and imidacloprid on honey bee colonies (*Apis mellifera*). *Ecosphere*, 9(8), e02378.

4. Appendices

Appendix 1. Protocol separating hazardous chemicals at WR (Radix building)

Appendix 2. Training how to deal with bee stings (WR version - We encourage the other partners to develop their own training or safety protocol if they do not have one yet).

Appendix 1. Protocol separating hazardous chemicals at WR (Radix building)

Separating hazardous waste at Radix



SUBSTANCES/ SOLUTIONS		Waste number	EXAMPLES	PACKAGE
HIGHLY TOXIC 05	LIQUID>	10CA20247660	Solutions of chemicals which are carcinogenic, reprotoxic, teratogenic, mutagenic, skin-penetrating. For instance: heavy metals, biocides, DMF, acetamide, phenol, chloroform, formaldehyde, formamide, acrylamide, mineral oil, solid ethylenedibromide, 2-mercapto-ethanol, glutaraldehyde	PE can (for liquids)
	SOLID>	10CA20247609		PE barrel (for solids)
HIGHLY REACTIVE 06	LIQUID>	10CA20247660	Chemicals with one of the following characteristics should be kept separately from all other chemicals: (not packed in PE-barrels): explosive, highly flammable, highly oxidizing, highly corrosive, burning with water, polymerizing. Example: F, Mg, Na, K, C ₃ -carbide, H ₂ O ₂ , (ge)chlorine, pyridine, chlorine, nitric, permanganate, org. peroxide, oleum, POC _{L3} , SOC _{L3} , mycotoxins, nitric acids and nitrate, cyanides, phosphor (concentrations)	Connet \$5800
	SOLID>	10CA20247609		Keep in original package if possible
ORGANIC, HALOGENS HIGH (≥5 % Br, Cl, F) 25		10CA20247648	Solvents with >5% (v/v) bromine, chlorine, iodide, fluoride > 5%. For instance: TCE, PER, dichloromethane	PE can / Metal can
	ORGANIC, HALOGENS LOW (<5% halogens flammable) Hazard cat. III	10CA20247608	Solvents with high conc. of methanol, ethanol, xylene, (di-ethyl)ether, acetone, hexanol, butanol, propanol (Organic oxygen mix). But <5% (low conc. halogens).	PE can (for liquids) PE barrel (for solids)
ANORGANIC BASE 41		10CA20247623	KOH, NaOH, Ca(OH) ₂ . If heavy metal concentration > 5%, write on label (with pack list instead of original bottles) No: cyanides	PE can (for liquids) PE barrel (for solids)
ANORGANIC ACID 31		10CA20247658	Sulfuric-acid, chloric-acid, phosphoric-acid. If heavy metal concentration > 5%, write on label (with pack list) No: nitric-acid → Nitric acid write on label Code 3F.	PE can (for liquids) PE barrel (for solids)
ORGANIC ACIDS		10CA20247619	Weak organic acids e.g. acetic acid, oxalic acid, fumaric acid, trichloroacetic acid	PE can (for liquids) PE barrel (for solids)
PHOTO DEVELOPER 44		10CA20247616	Silver, heavy metal containing mixtures	PE can
MERCURY, METALLIC 79		10CA20247622	Thermometers, if mercury is escaping (incident): obtain safety rules, call 88112/ \$5800 Conditions: double packed with absorbent Call \$5800	PE barrel = absorbent
ANORGANIC SALTS 04		Slitk	Mind the exceptions! Always ask your lab supervisor. E.g. Na, K, Ca, Mg, Li, salts / Sodium, Potassium, Calcium, Magnesium, Lithium salts	Slitk PE can (for liquids) PE barrel (for solids)
OBJECTS				
BOROSILICATEGLASS AND CHEM. CONTAMINATED BOTTLES, FLASKS, JARS 08A		Waste number	EXAMPLES	PACKAGE
	CLEAN GLASS BOTTLES AND JARS	10CA20247612	Glass-tubes, Erlenmeyer-flasks, pipettes, contaminated jar and bottles, Scott Duran bottles. No: Pipette tips contaminated with highly toxic and highly reactive chemicals. Keep attention for instructions on the yellow waste container	YELLOW 240 I container (in corridors)
BATTERIES (< 1 kg / piece)		Glass bins	Bottles (ethanol, methanol, chloroform, hexane, acetone, pentane, ether) Rinse bottles from acid/ base flasks. Cleaning should be easy (evaporating or rinsing with water). Make sure org. label is illegible! Keep attention for instructions on the brown waste container E.g. penlight's, button cells, photo camera cells. Exception: 100% Lithium acc's, (<250 g/po) collect separately → call \$5800	(BROWN) 240 I container (in corridors) PE barrel/ KCA box
	PACKAGING with lab chemicals residue	10CA20247647	Plastics and metals. No: lab glass and Biohazard HEPA filters from Biosafety cabinets class II from BSL-II and BSL-III labs	PE barrel/double packed
FILTERS with lab chemical residue 01E		10CA20247624	Used active charcoal	PE-barrel
NEEDLES, SCALPELS, OTHER SHARPS		10CA20247617	Used in association with Quarantine, human pathogens, GMO's (contaminated or not with Biological Agents) (yellow sharps container)	Needle box/ biohazard
OFFICE WASTE (incl. small batteries) 82		10CA20247618	Ink, glue, cartridges, correction liquid, PC-cleaner, printer cartridges, batteries.	PE barrel / small toxic office waste box (in corridors)
MERCURY CONTAINING OBJECTS 79		10CA20247613	Thermometers. Offer to Facility Services: Plant \$5800 (Offer in air-tight / air-locked bottles:)	PE barrel
TL-LAMPS, long tubes/standard sizes		10CA20247661	Long tubes in special lamp boxes: Offer to Facility Services: Plant \$5800	In solid box
EXPLANATION				

	Severe health hazards		Health hazards		Acute toxicity
	Explosive		Flammable		Oxidising
	Corrosive		Gases under pressure		Environmental hazard

Please fill out the label on the barrel. Use the correct waste number. You can order new barrels and labels at Facility Desk, phone: 86666

- * Highly reactive waste should be collected and packed separately from any other waste.
- Please dilute acid- or base solutions > 10 N to a concentration < 10 N before disposal, by adding acid/ base to water
- Please close and clean full labeled barrels and put them in the corridor outside the lab. Facility Services will collect them. Please don't overload barrels.
- Info: Facility Desk 86666. Report accidents & incidents via alarm number 88112

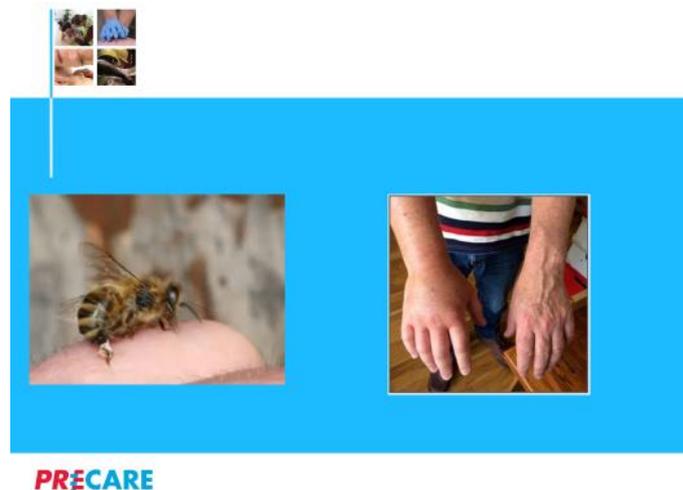
Proper separation of waste is sustainable, cheaper, safer for human/animal and better for the environment!!

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Appendix 2. Training how to deal with bee stings (WR version - We encourage the other partners to develop their own training or safety protocol if they do not have one yet).

Training how to deal with bee stings

19.08.2019



Stings :

- Remove the stinger as soon as possible
- If possible, remove poison (only beneficial against itches as poison is already in the body)
- Cool the place where you have been stung
- If necessary, use a cream for insect stings and -bites
- Sting in face > swelling, we use our own anti-histamine medication pills prescribed by doctor

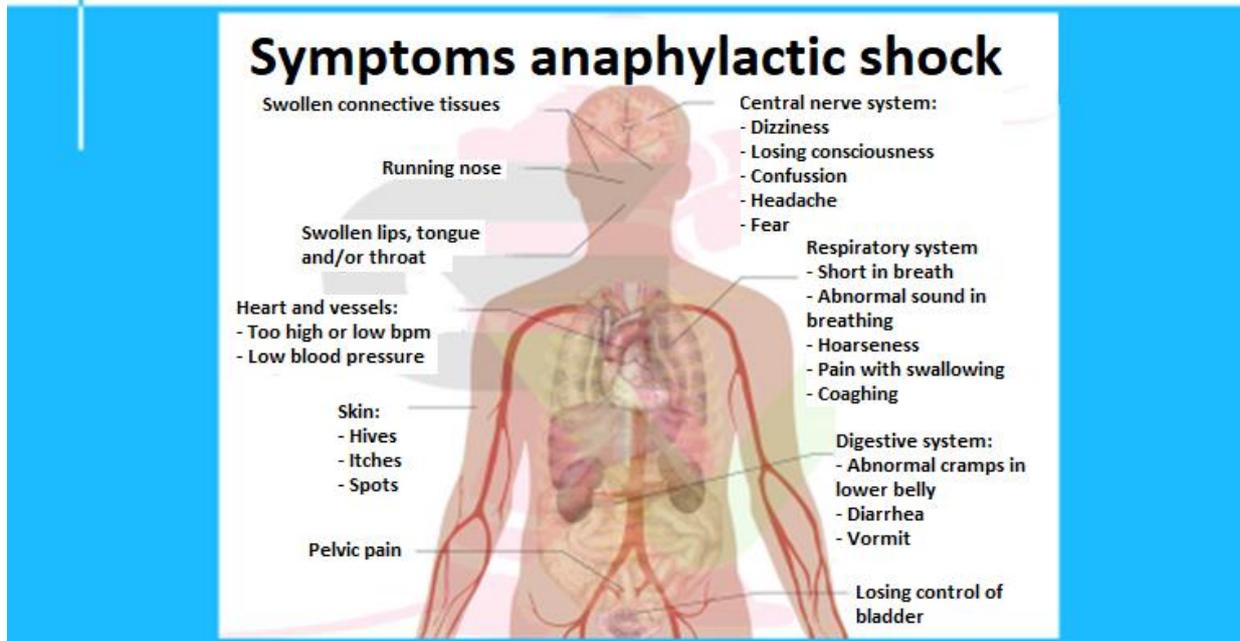


What to do?

- In doubt always call 112! If it is not as bad as you thought, they will only give you advice. If it is worse, then they will instruct you and possibly send an ambulance.
- 112 knows your location when you are carrying an android phone, but be sure to know it yourself! It saves the workers at 112 the time they need to track your location.
- If a bee stings you in the mouth, always call 112 for help!

Signs of an anaphylactic shock:

- Itches, red spots, swollen connective tissue, swollen mouth and lips, a running nose, low blood pressure, losing consciousness, difficulty breathing, cramps and urinating.
- In the beginning hardly any symptoms will show!



PRE CARE

Process of anaphylactic shock:

1. Your blood pressure drops: your body is removing blood from organs, starting with the skin, which becomes cold and clammy/sweaty hands
2. Your body is removing blood from muscles: you cannot control your bladder and may need to urinate or want to urinate
3. Your body is removing blood from your digestive system: you are getting very thirsty or hungry. Do not eat or drink as this might speed up the whole shock process!
4. Your body is removing blood from your remaining organs: you are getting confused, even respond aggressively and you might lose consciousness
5. Death will follow if no treatment is given during the previous steps



Important emergency equipment for the field:

- Emergency blanket (gold/silver)
- Chemical cooling pack
- EpiPen and anti-histamine (preferably matching yellow cover)
- Highly visible safety jacket

In case of emergency:

- First bring yourself into safety
- Make sure that the surrounding as well as the people around you are safe
- THEN take the victim to safety
- Make sure that the victim is responding!
- Have both your hands available while being on the phone with 112
- Always call 112 in case of doubt, 112 can be called for advice, emergency and ambulance
- Watch the victim's breathing; this can be done by the chin lift
- When a victim breaths at least 2-3 times every 10 seconds put him/her on their side
- When a victim breaths less than 2-3 times every 10 seconds, then CPR is necessary!