EFFECTS OF EARLY AND REPEATED EXPOSURE TO BOSCALID ON *APIS MELLIFERA*

ECOPHYTO II+

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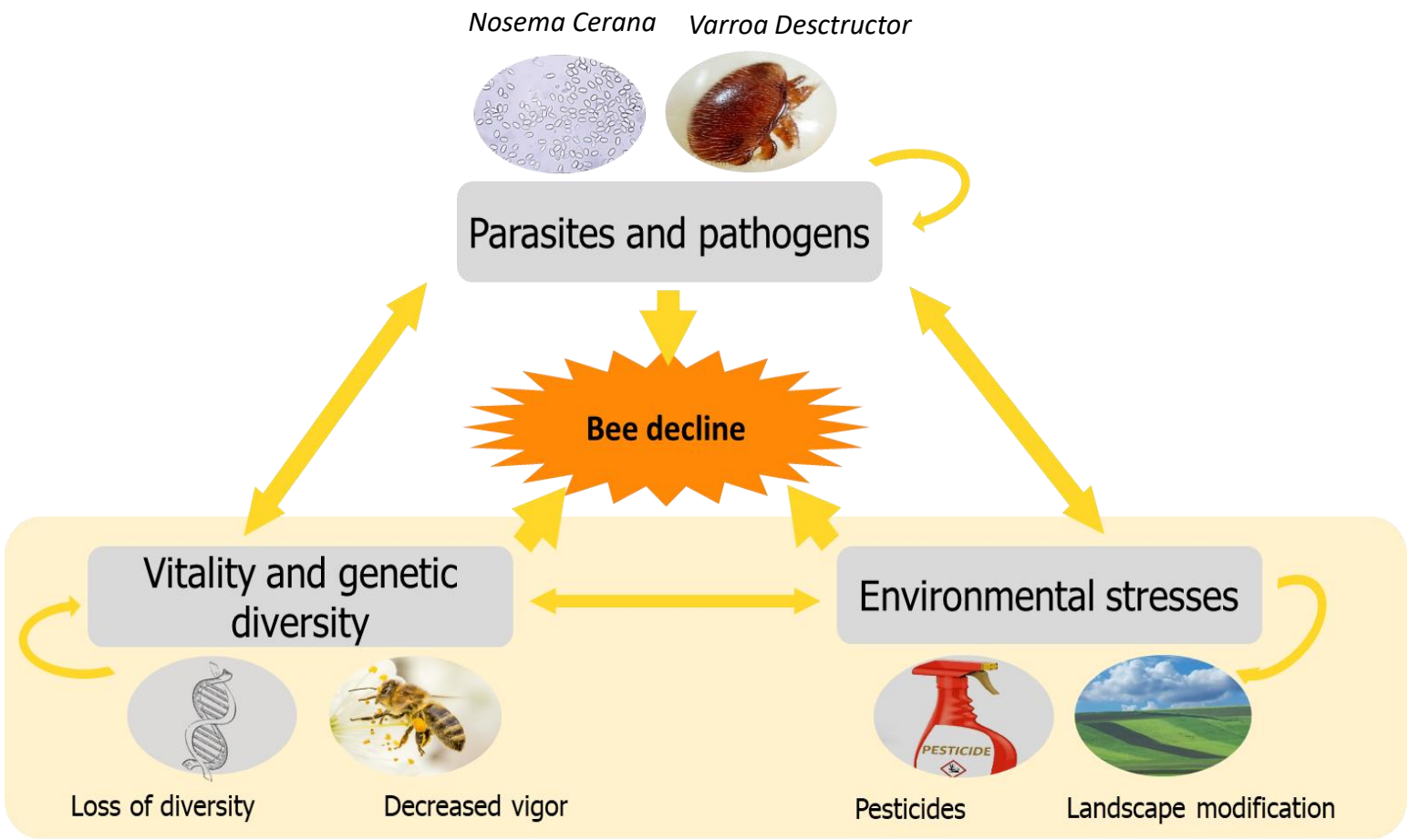
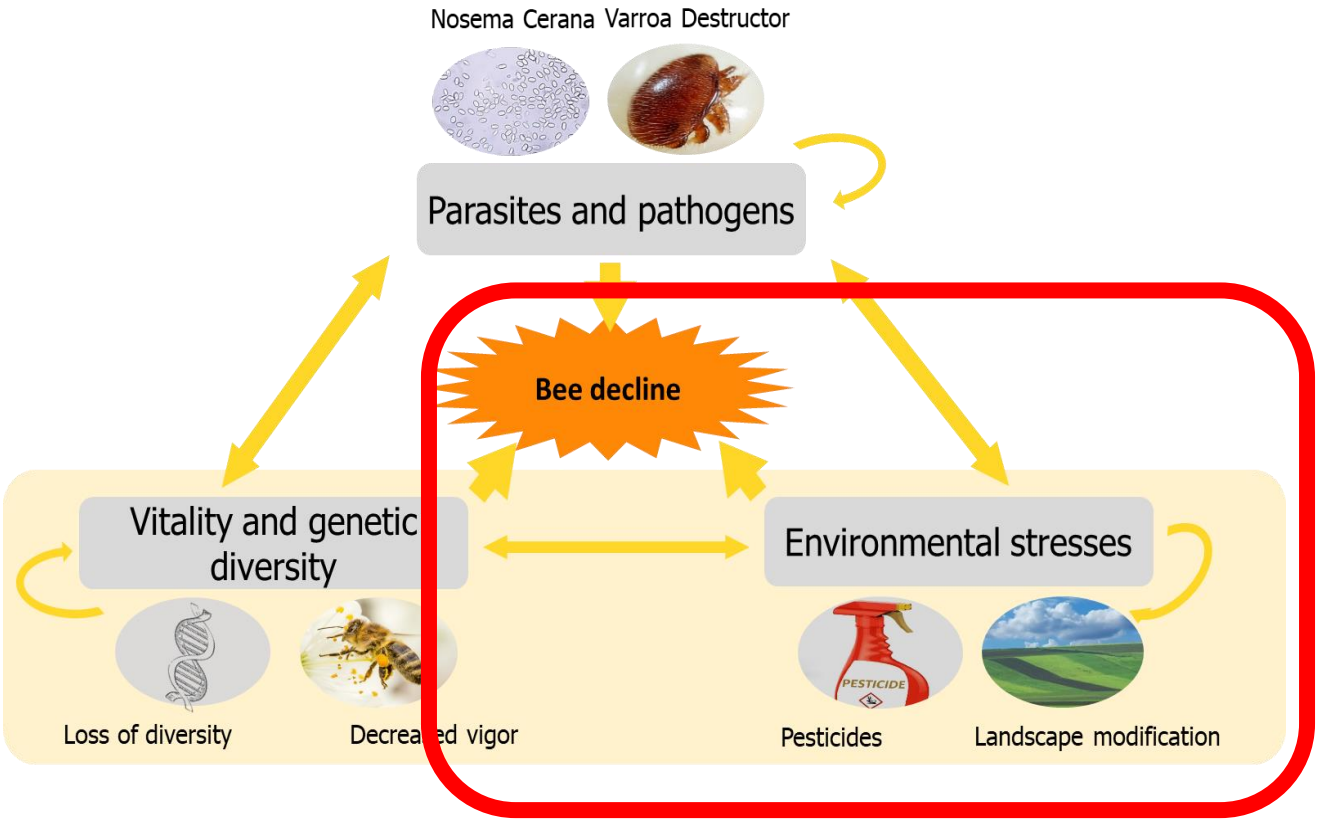
19 - 09 - 2022

Bee decline: a multifactorial cause



Sánchez-Bayo *et al.*, *Biol Cons* (2019)
Müller *et al.*, *Basic and Applied Ecology* (2018)
Goulson *et al.*, *Science* (2015)
Potts *et al.*, *Trends in Ecology & Evolution* (2010)

Pesticides



- Pesticides cause adverse lethal, sublethal and combined effects on bees
- Sublethal or combined effects of pesticides are not well documented
- The sublethal effects of pesticides on non-target species are complex to study and likely underestimated.

Sánchez-Bayo *et al.*, *Biol Cons* (2019)
Müller *et al.*, *Basic and Applied Ecology* (2018)
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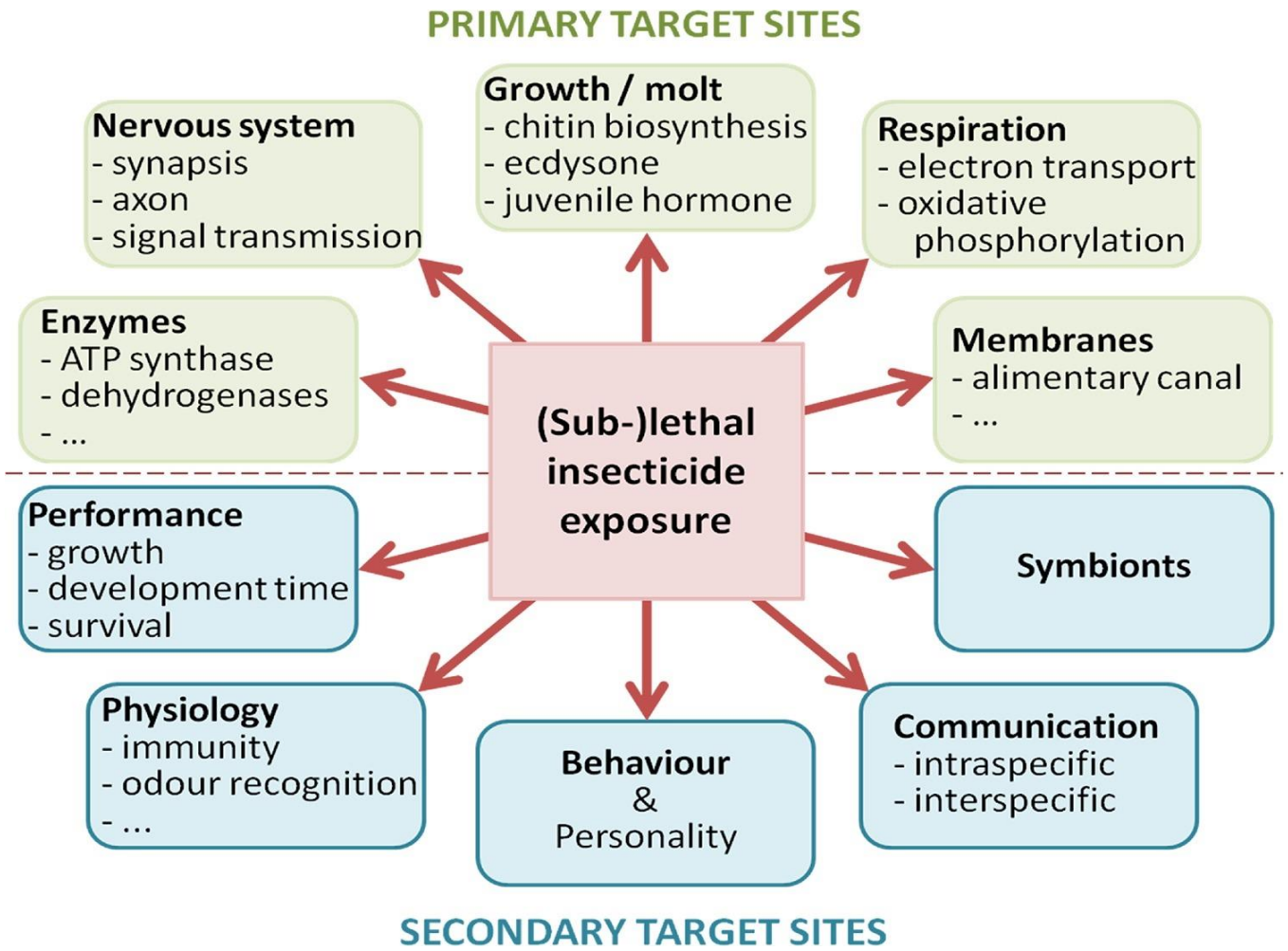
Sublethal effects of pesticides



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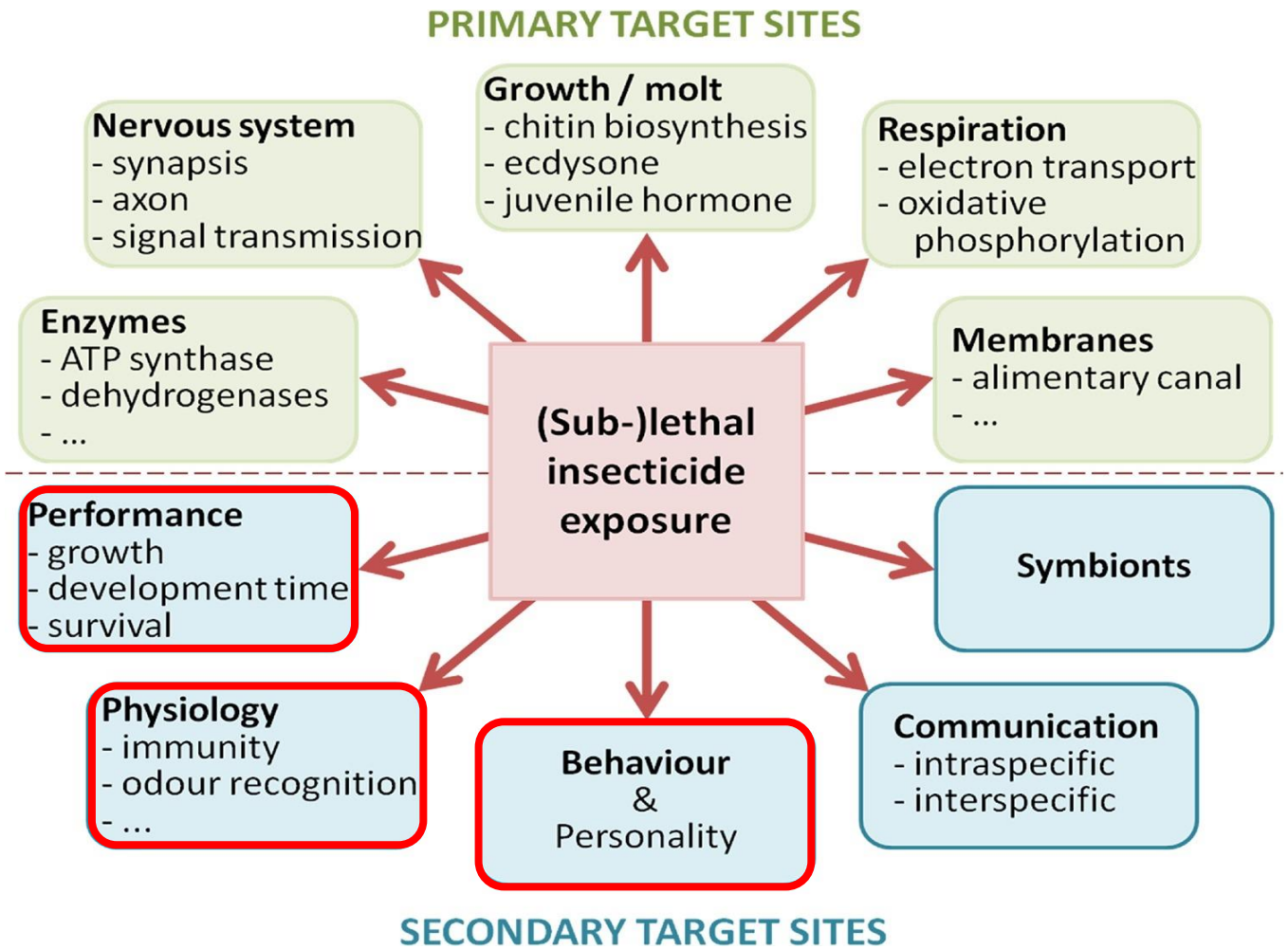
Effects of pesticides on individuals

Sublethal effects of pesticides



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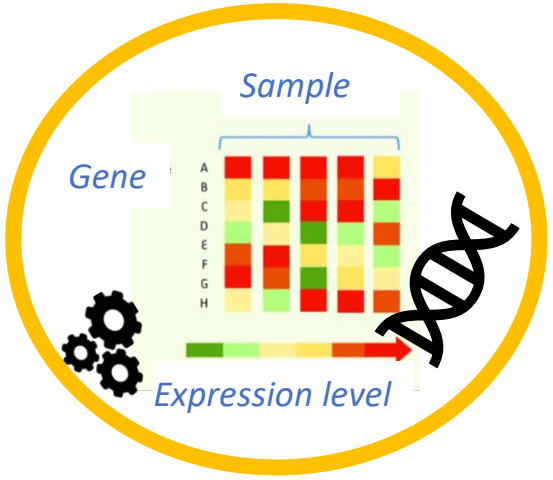
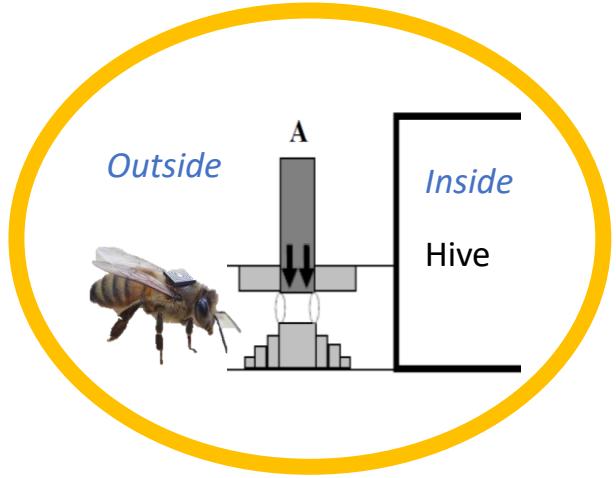
Effects of pesticides on individuals

ECOPHYTO 2+
EXPLORA
Impact of larval exposure on the life history of *Apis mellifera*





Analysis of the expression of immune/detoxification genes and chemical compounds released by individuals

Activity and behaviour analysis with a RFID system

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PI: F-J RICHARD

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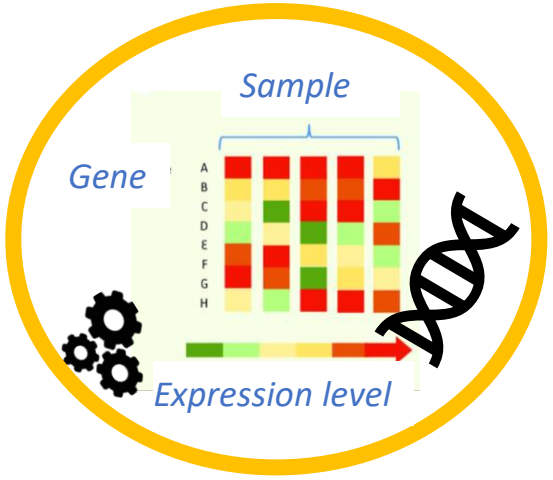
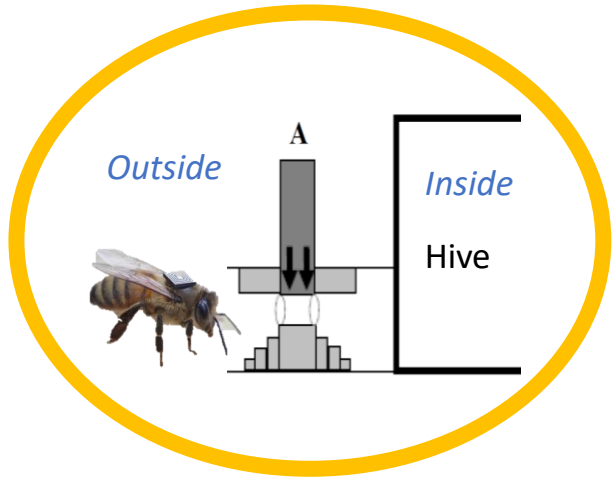
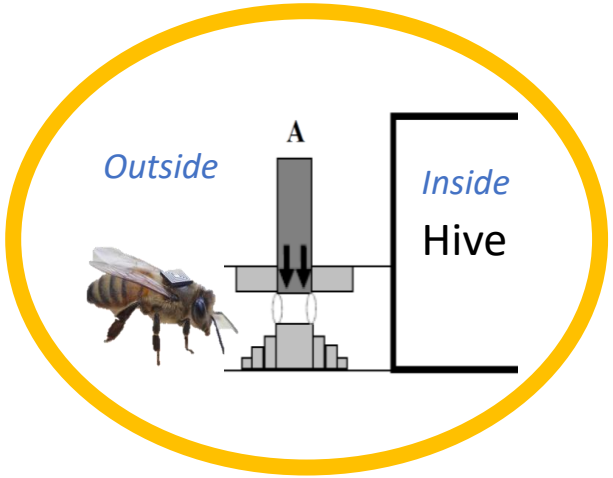


Analysis of the expression of immune/detoxification genes and chemical compounds released by individuals



Activity and behaviour analysis with a RFID system

What are the effects of early and repeated exposure of Boscalid on *Apis mellifera*?



Impact of a fungicide

Boscalid: Fungicide

- Carboxamide family
- Inhibits ATP production (blocks ATP activity of enzyme the succinate dehydrogenase)

- Against brown rot, gray mold...
- In the orchards, on rapeseed...
- High dose (high LD50)
- In many commercial solutions (Pristine®, Pictor Pro ®, Cantus ® ....)

Modality of intoxication

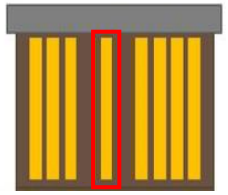
Dose of intoxication → Representative dose of the one found in the environment

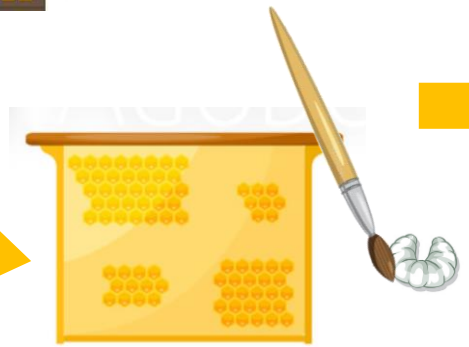
→ Oral exposition

→ **Protocol developed by INRAE and adopted at the OECD**

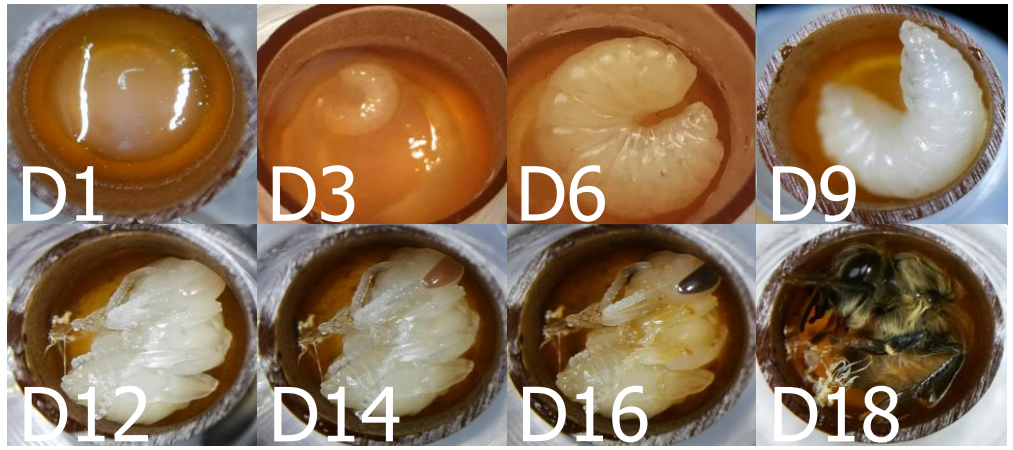
Böhme *et al., PLOS ONE* (2018)
Böhme *et al., Jour. Api. Rech* (2017)

Larval rearing protocol (OECD)

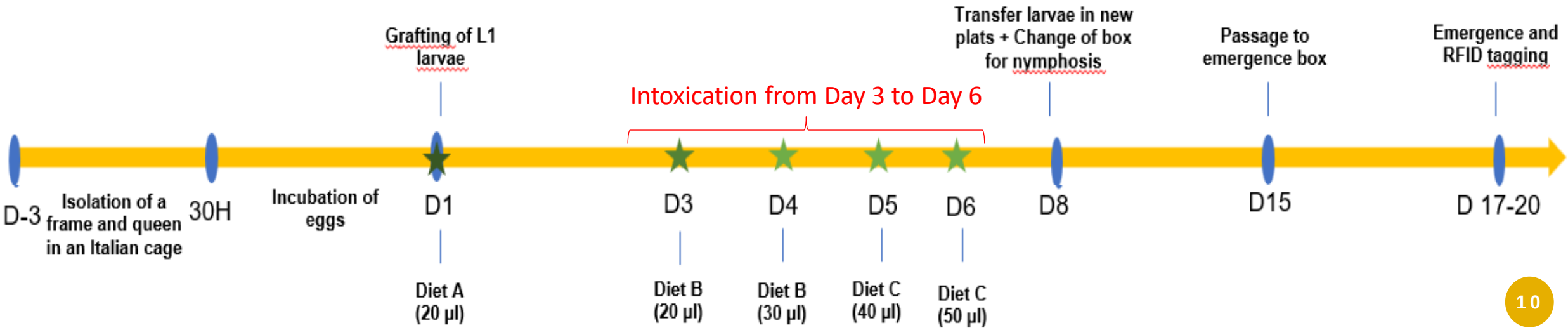




Intoxication from Day 3 to Day 6



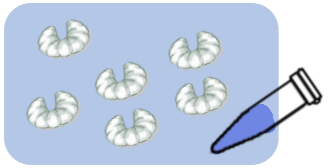
RFID



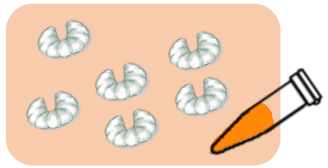
Experimental approach

Chronic exposure in the larval phase

Control



Boscalid

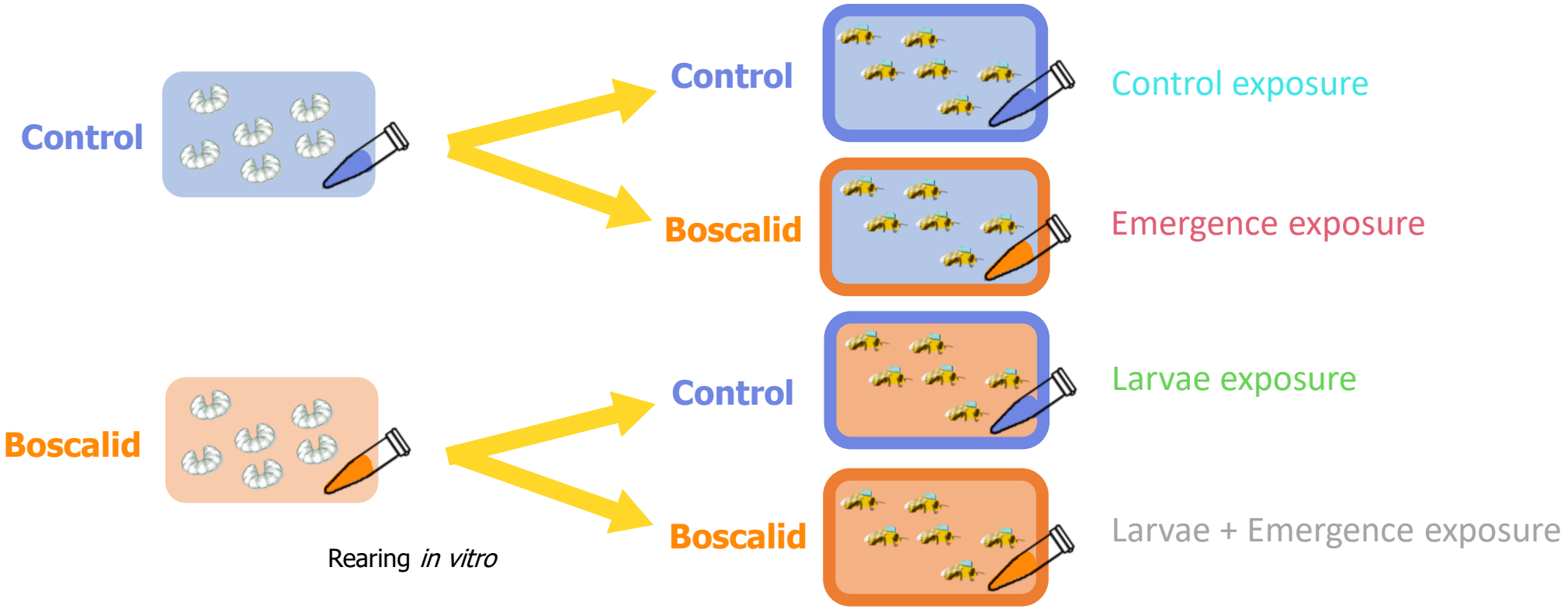


Rearing *in vitro*

Experimental approach

Chronic exposure in the larval phase

Exposure 48h after emergence

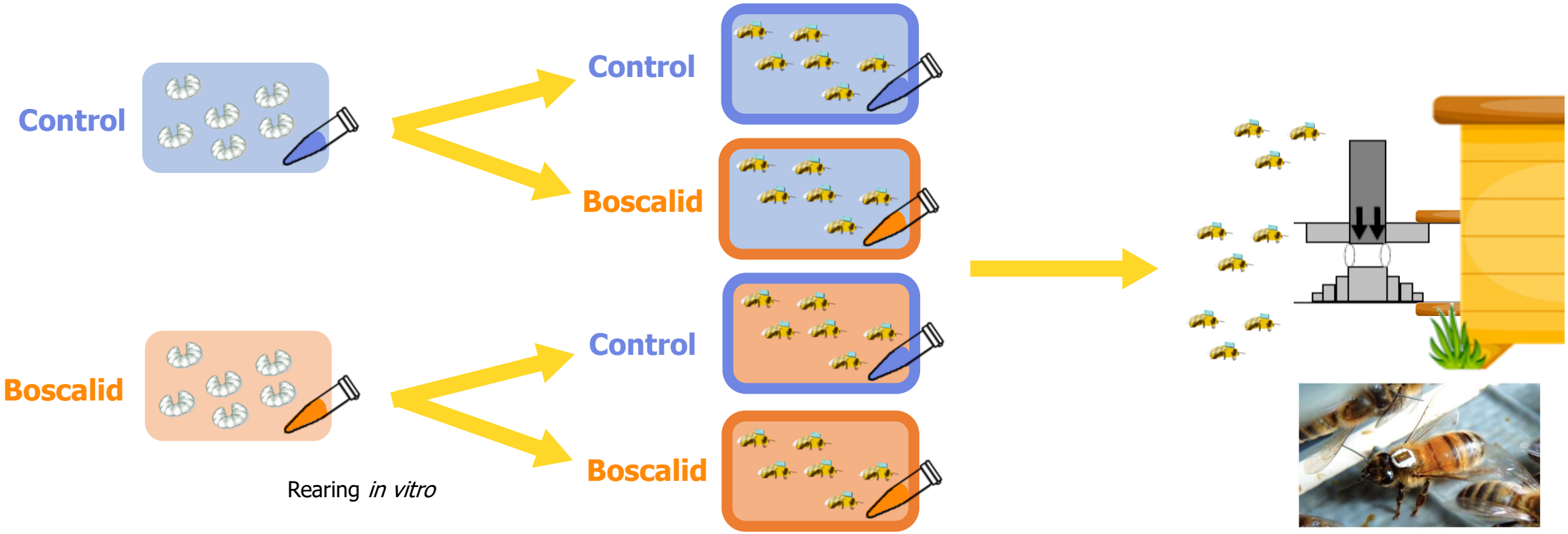


Experimental approach

Chronic exposure in the larval phase

Exposure 48h after emergence

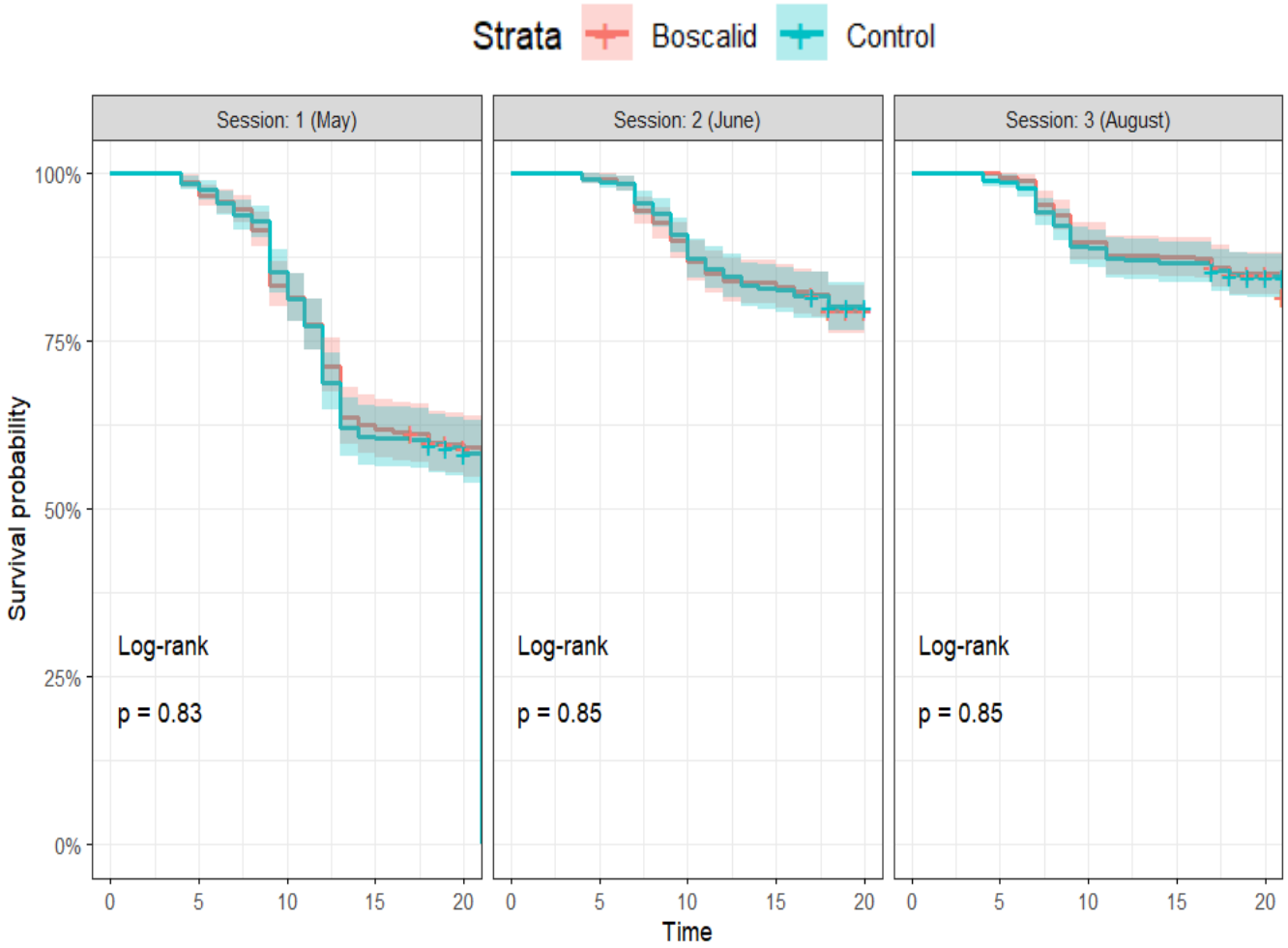
Released into the RFID hive



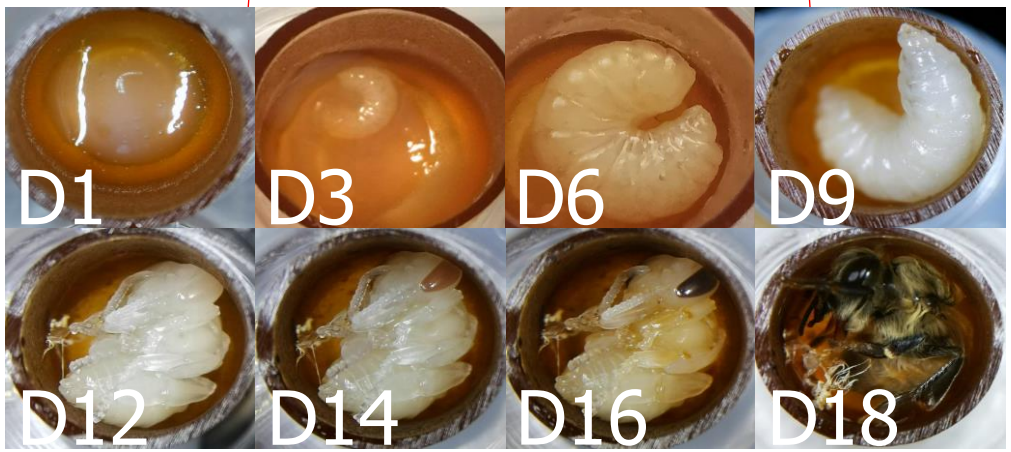
Bee with a RFID chip

- Three replicas carried out (May, June and August 2021)
- Taking into account seasonal variations
- 1073 individuals tracked in RFID

Larval mortality in laboratory



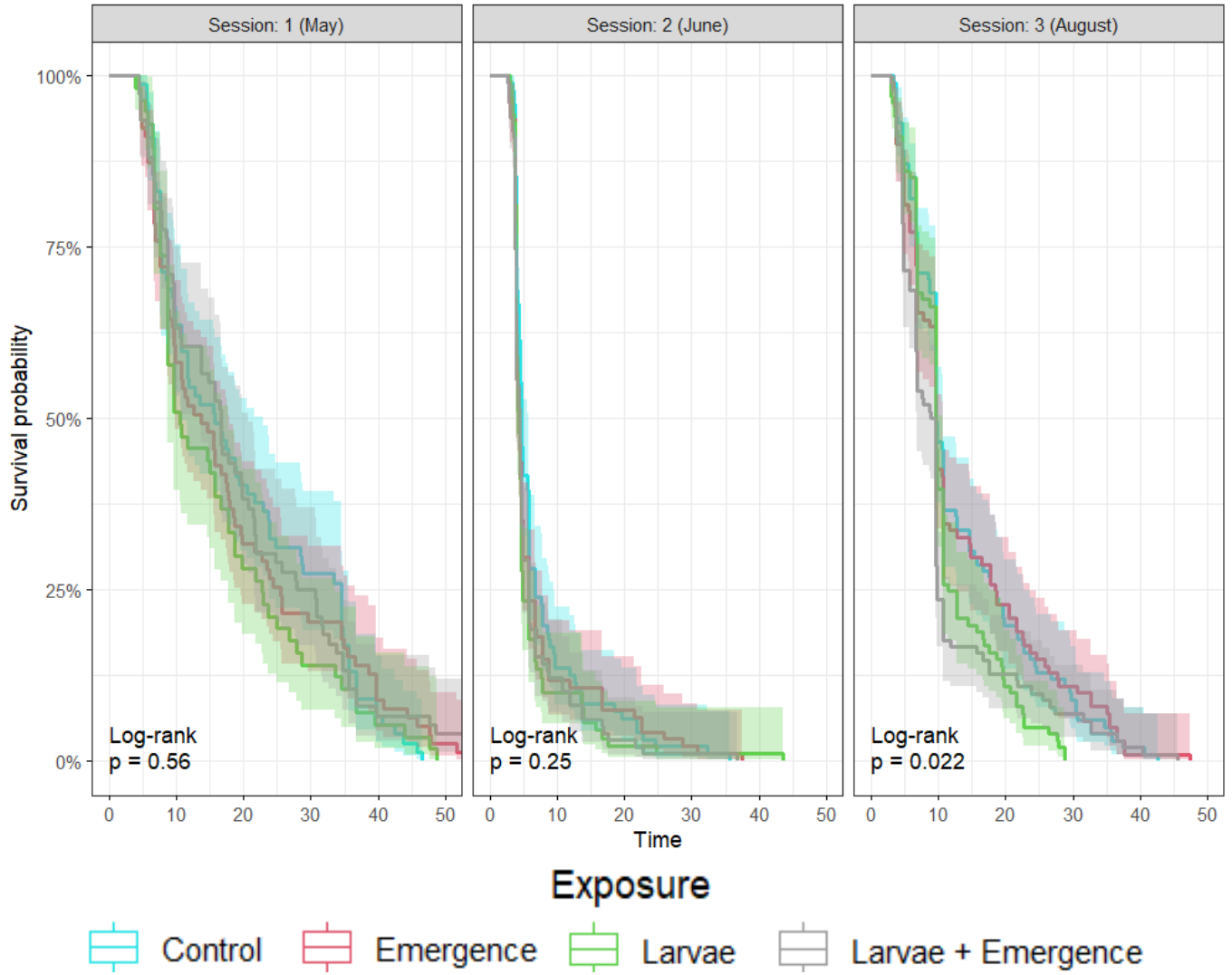
Intoxication from Day 3 to Day 6



- No direct effects on larval mortality
- No effect on emergence of bees in laboratory rearing
- The first session had a high overall mortality which allowed the protocol to be improved

Survival of bees *in vitro* recurrently exposed to Boscalid

➔ Lifespan is shorter in session 2



Survival of bees *in vitro* recurrently exposed to Boscalid

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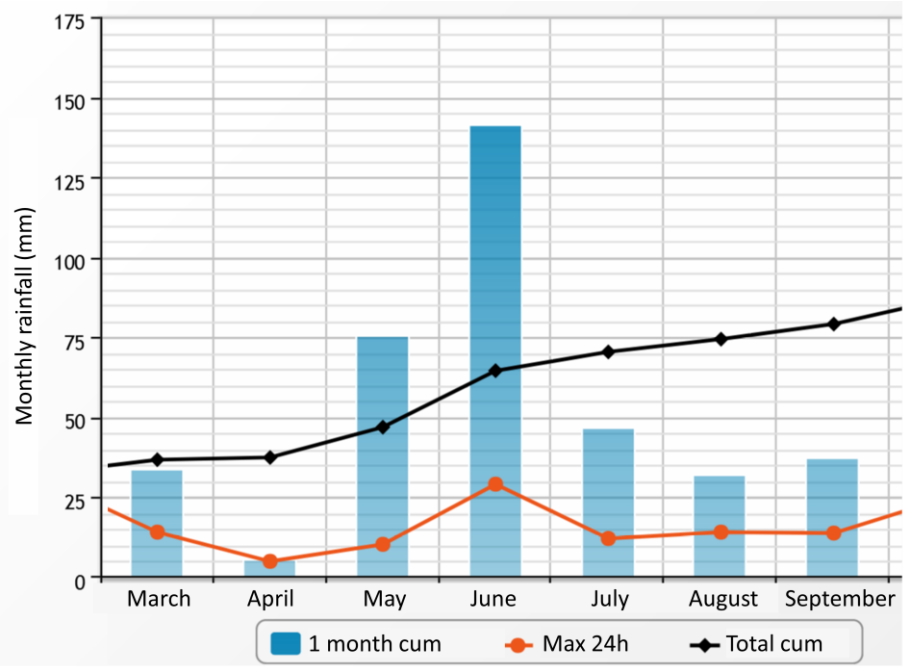
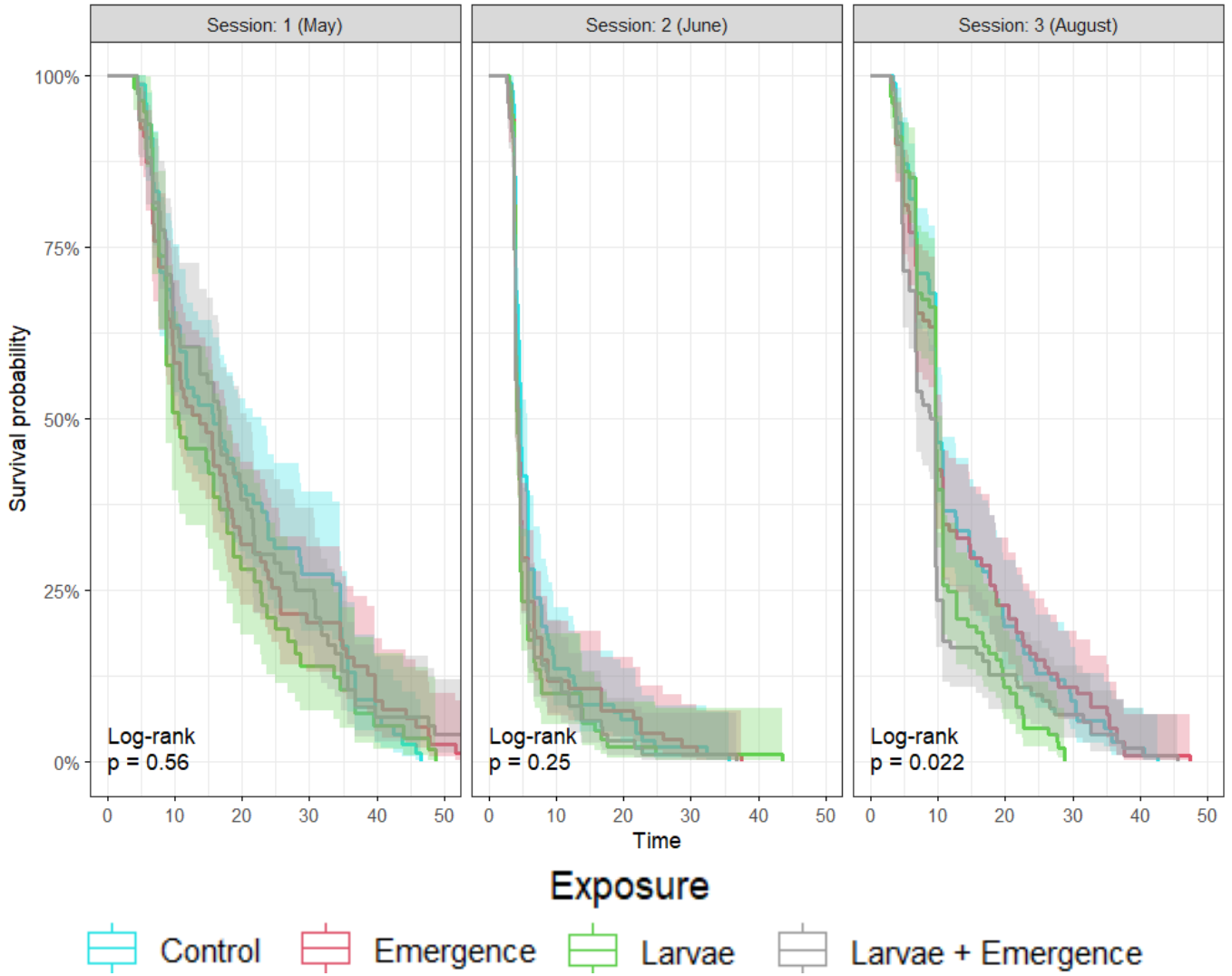
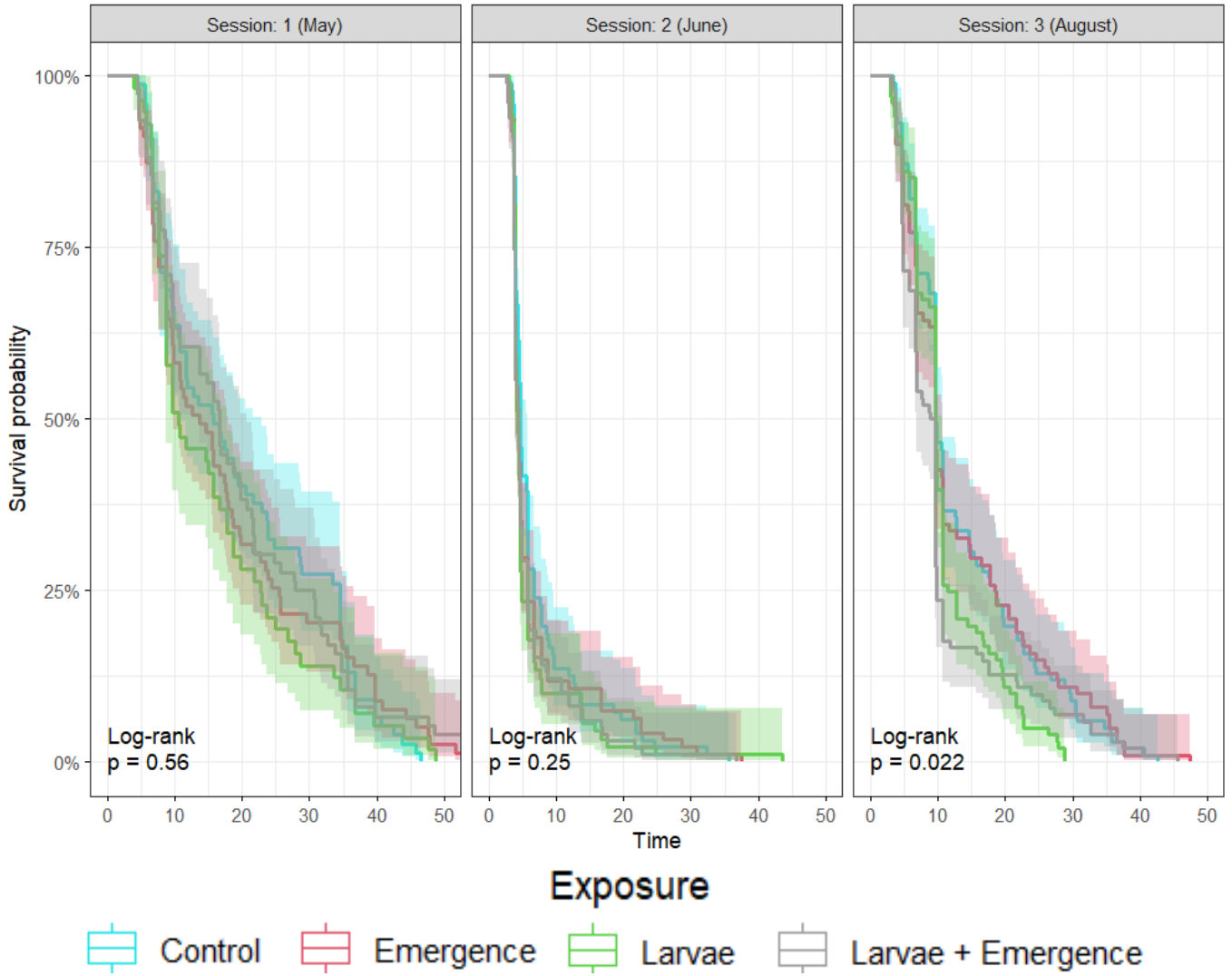


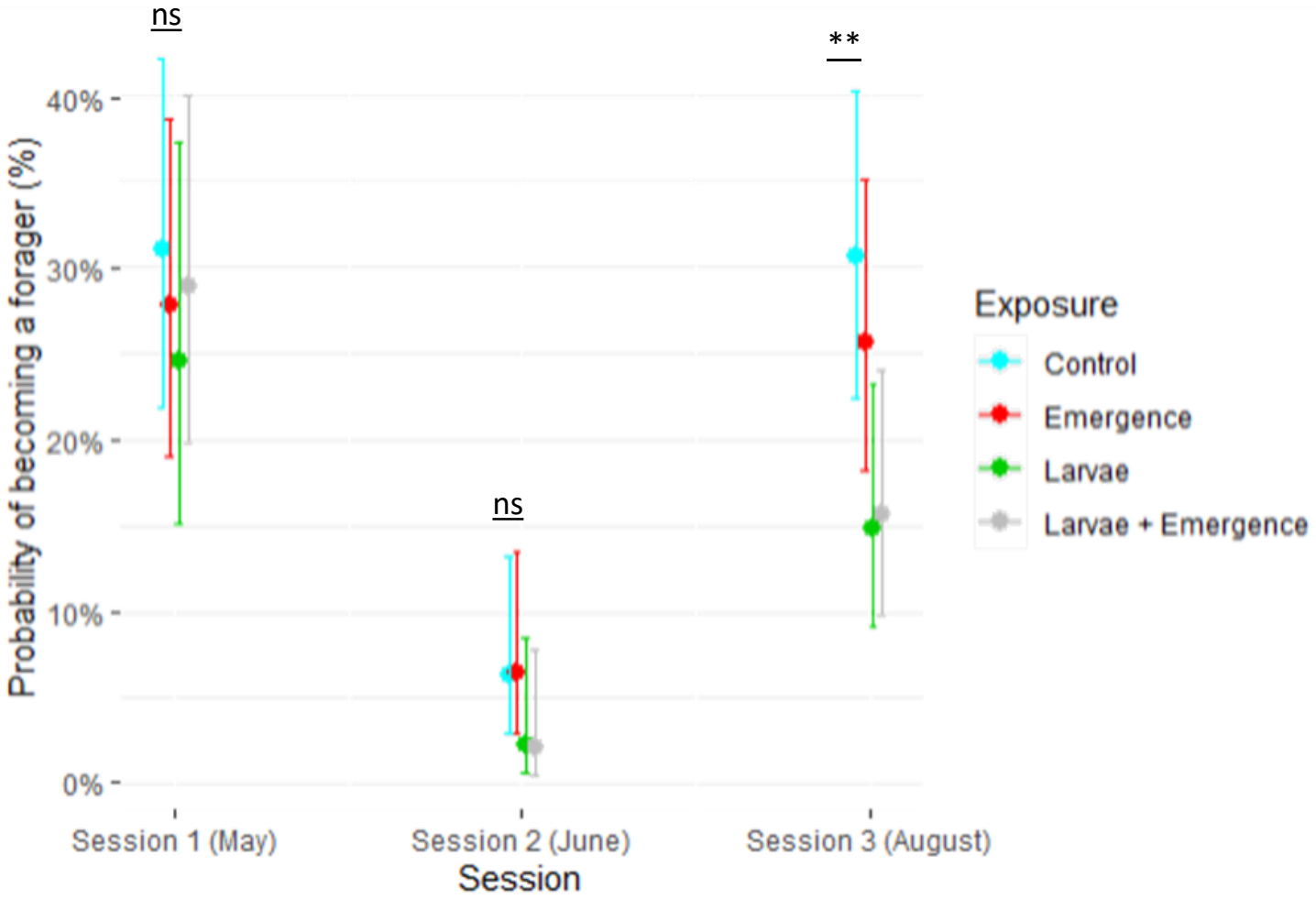


Survival of bees *in vitro* recurrently exposed to Boscalid

- Lifespan is shorter in session 2
- No difference between the exposure modalities for session 1 and 2
- Significant difference between control and Boscalid larvae exposure modalities for replicate 3

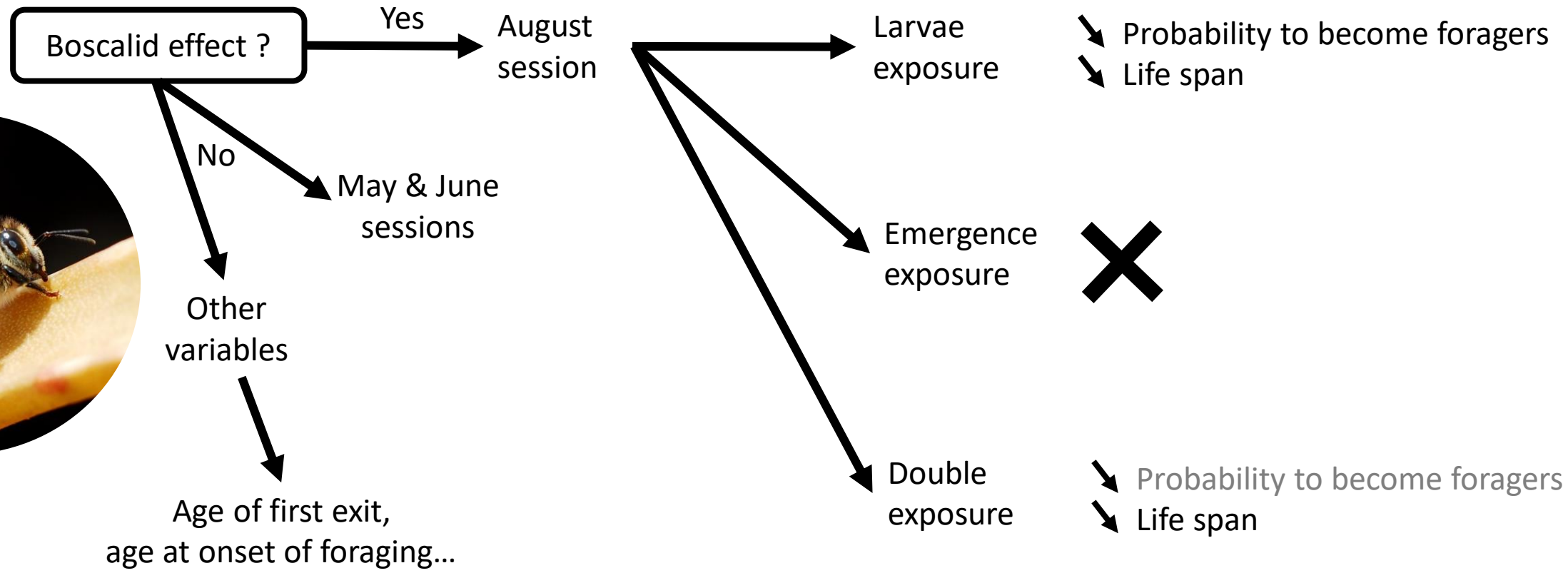


Probability of becoming a forager



- High effect between different sessions
- Significant effect of boscalid in session 3 in a larval exposure
- High tendency in a double exposure of boscalid

Effects of Boscalid on bees reared *in vitro* under *in situ* conditions

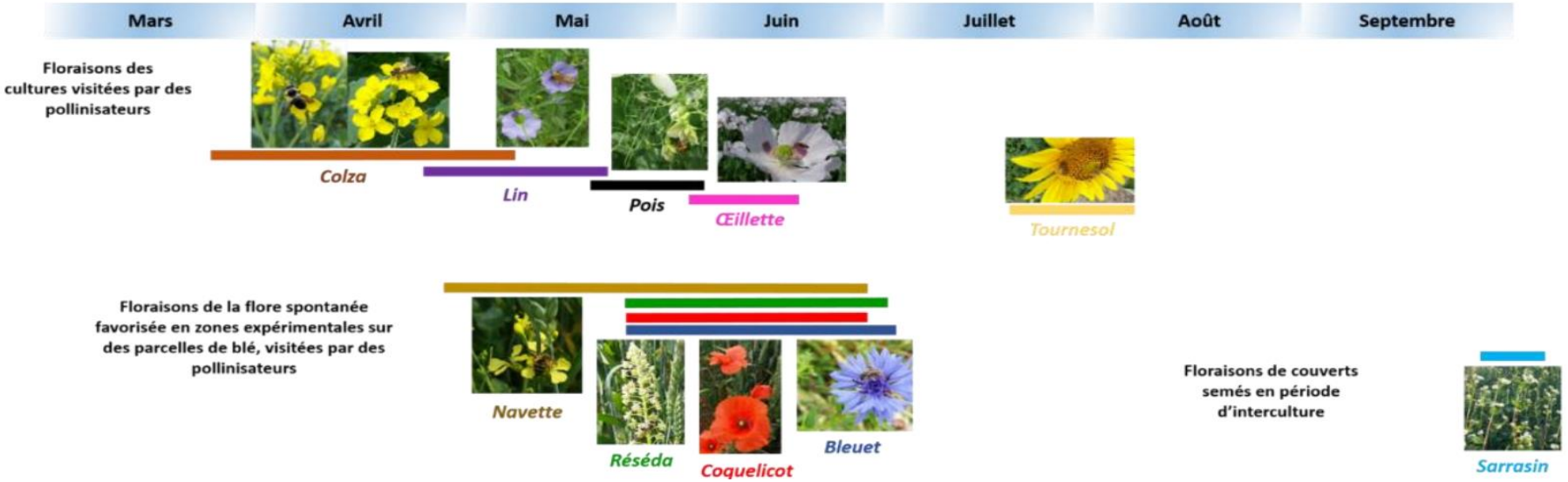


- The effects of pesticides are not constant throughout the season
- Boscalid impact life history of *in vitro* bee exposed in larval stage
- Double exposure to Boscalid does not seem to affect the fate of bees simply exposed

Towards a better understanding of the effects of pesticides

→ The bee is an organism with great plasticity and will adapt to its environment:

- Weather, overall hive population, honeyflow and dearth periods, presence of varroa mites... …



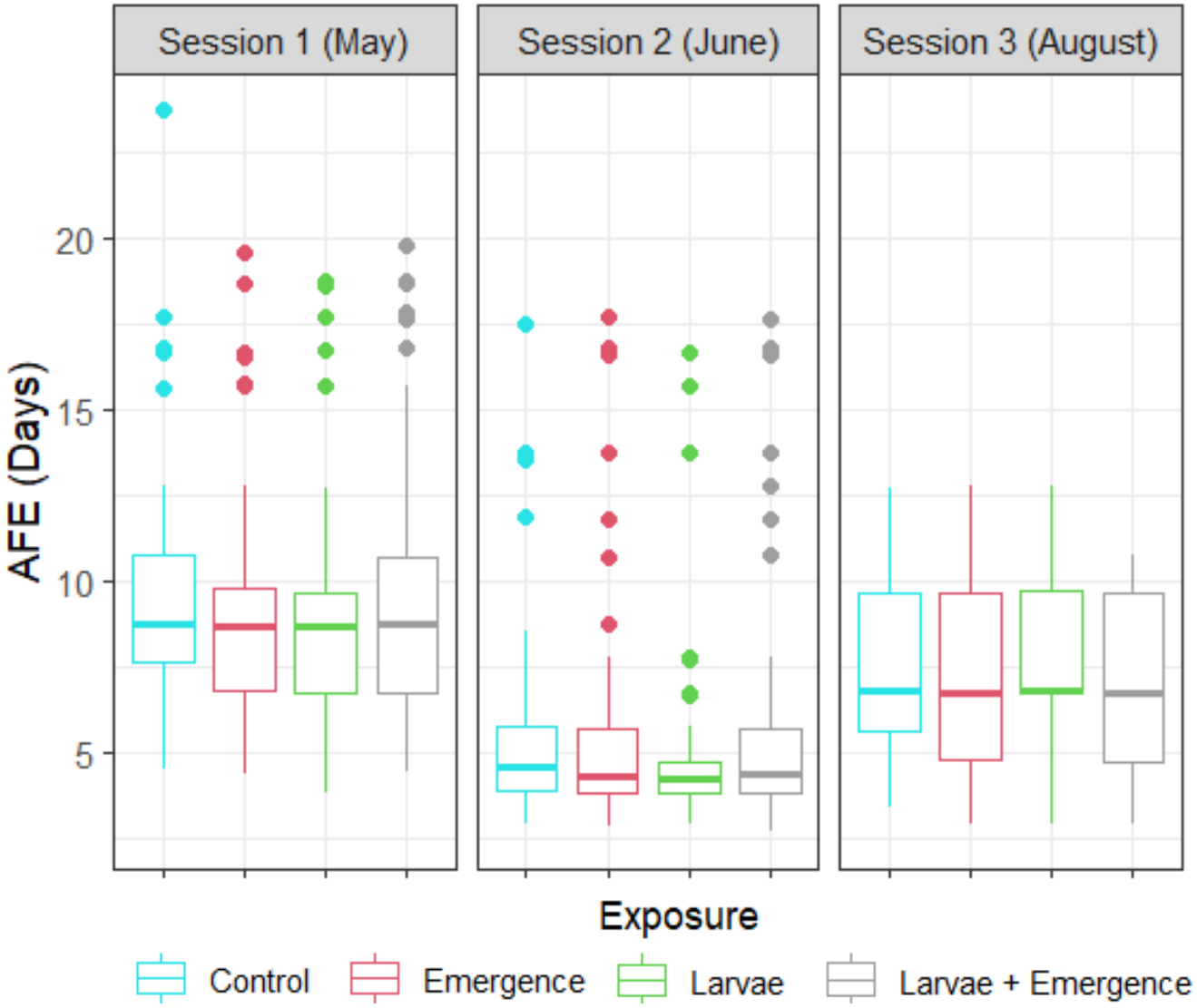
→ To generalize, the effects of pesticides in interaction with environmental stresses are probably under-evaluated.

→ It is important to look at the fate of bees exposed to the pesticide during the larval stage

Thanks to the team from INRAE du Magneraud and the University of Poitiers:

- Stephane Grateau
- Daniel Raboteau
- Carole Moreau-Vauzelle
- Maxime Pineaux
- Elisa Gomes
- Manon Desaivres
- Tiffany Laverre
- Pierrick Aupinel
- Fabrice Requier
- Freddie-Jeanne Richard

Age of first exit (AFE) of *In vitro* bees



- High differences between the different sessions
- No significant difference between the dates of first of the bees exposed to the different modalities

Effects of Boscalid on bees reared *in vitro* under *in situ* conditions

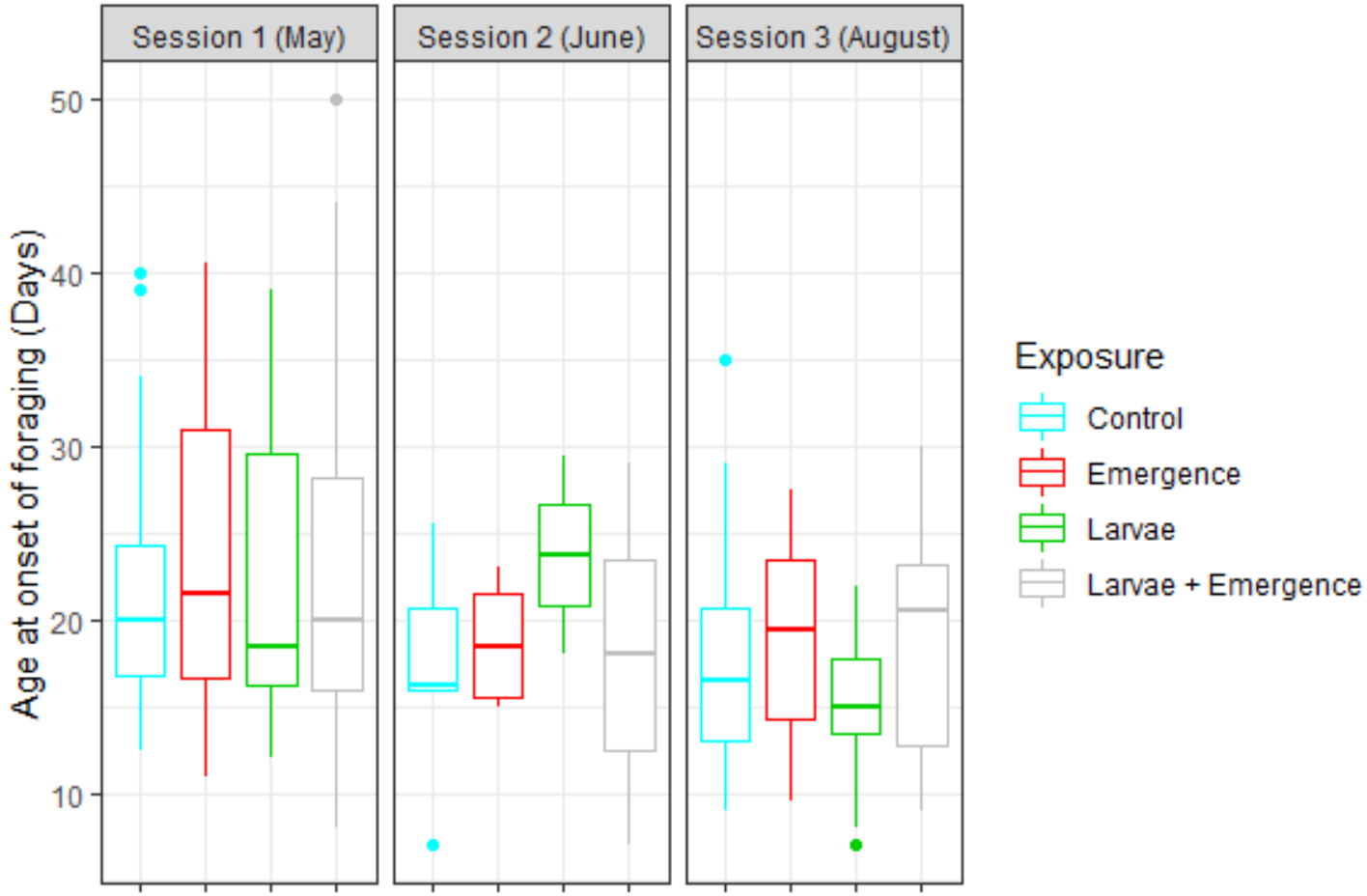
Synergistic effects of pesticides on the life cycle of bees

Variables	Session 1 (May)	Session 2 (June)	Session 3 (August)
Larval rearing	NS	NS	NS
AFE	NS	NS	NS
Longevity	NS	NS	Significant
foraging	NS	NS	Significant
AOF	NS	NS	NS

→ The effects of pesticides are not constant throughout the season

Age at onset of foraging

- No effect of pesticides on age of first foraging
- High variability in session 2 due to low number of foragers

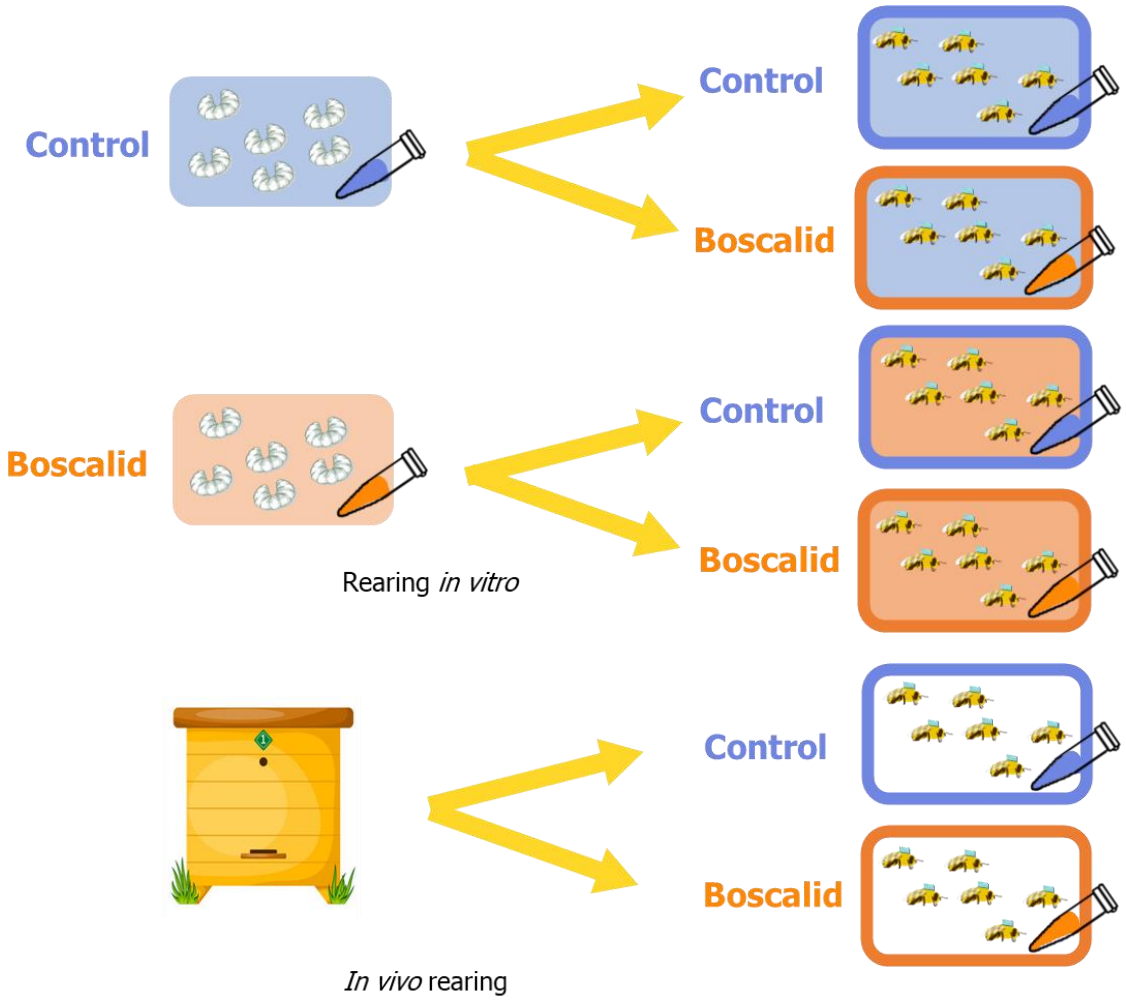


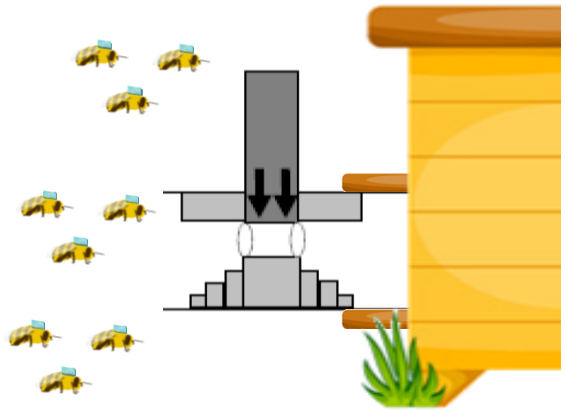
Experimental approach

Chronic exposure in the larval phase

Exposure 48h after emergence

Released into the RFID hive





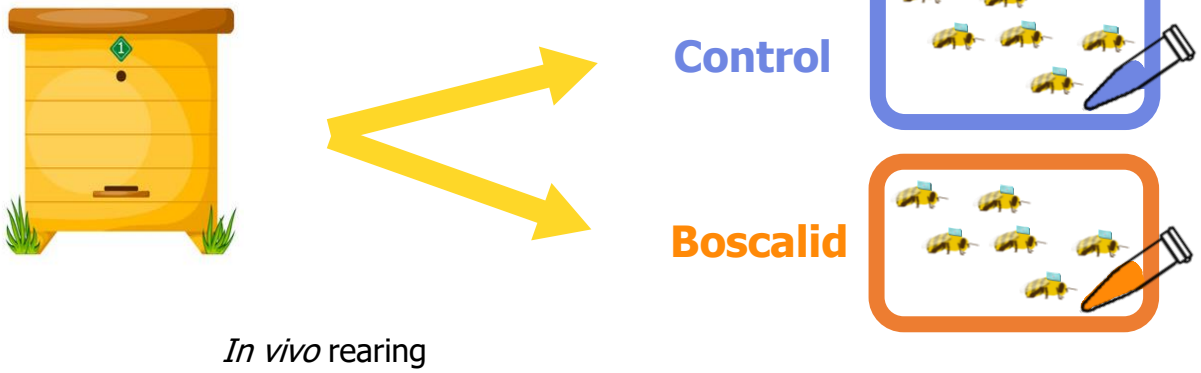
Bee with a RFID chip

Experimental approach

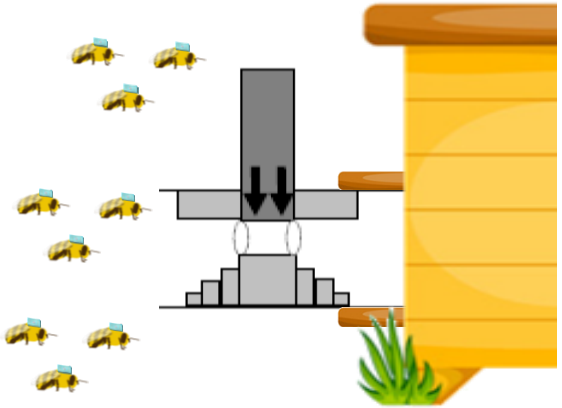
Chronic exposure in the larval phase

Exposure 48h after emergence

Released into the RFID hive



In vivo rearing

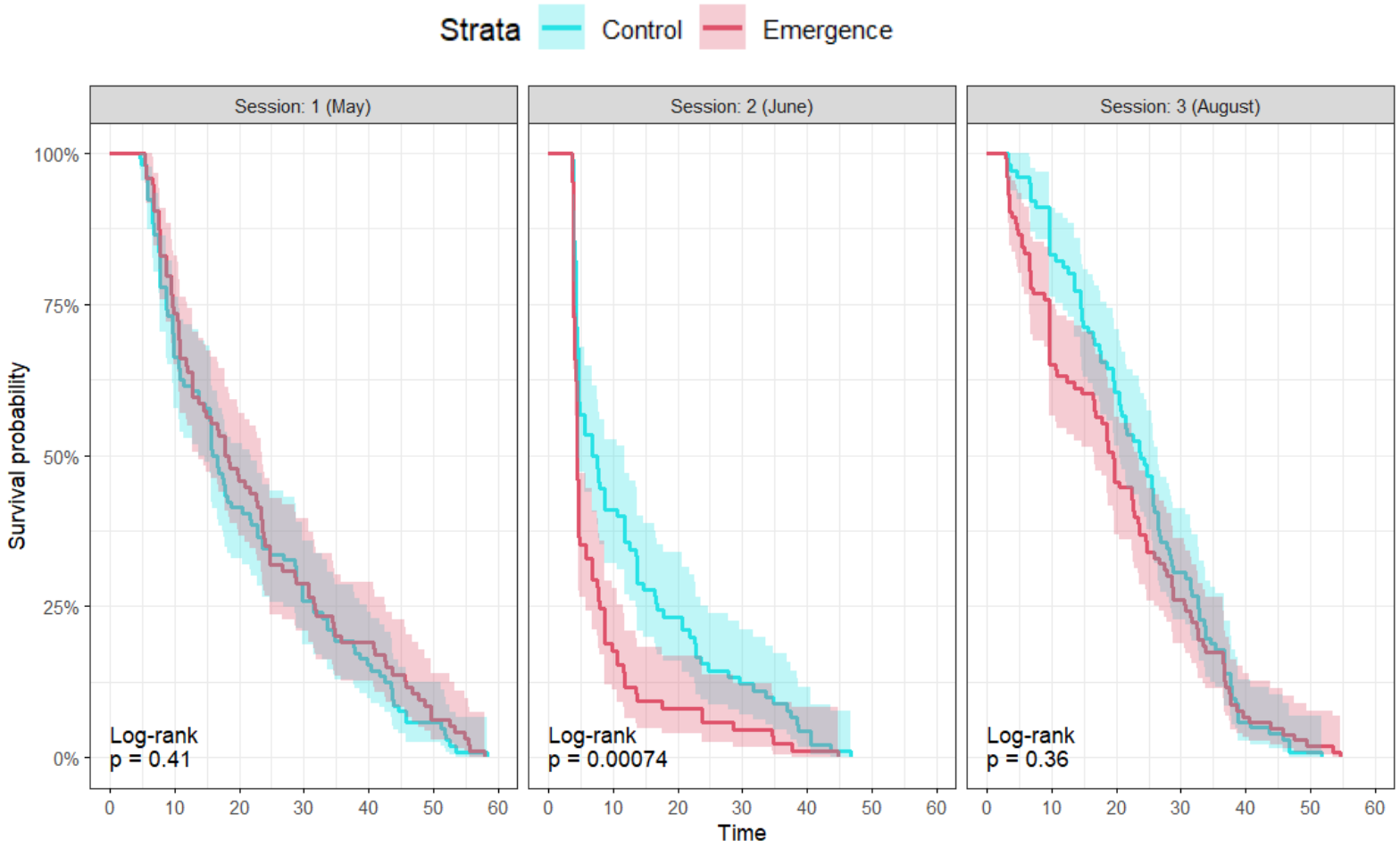


Bee with an RFID chip

- Three replicas carried out (May, June and August 2021)
- 1625 individuals tracked in RFID

Taking into account seasonal variations

Survival probably of *in vivo* bees exposed to emergence

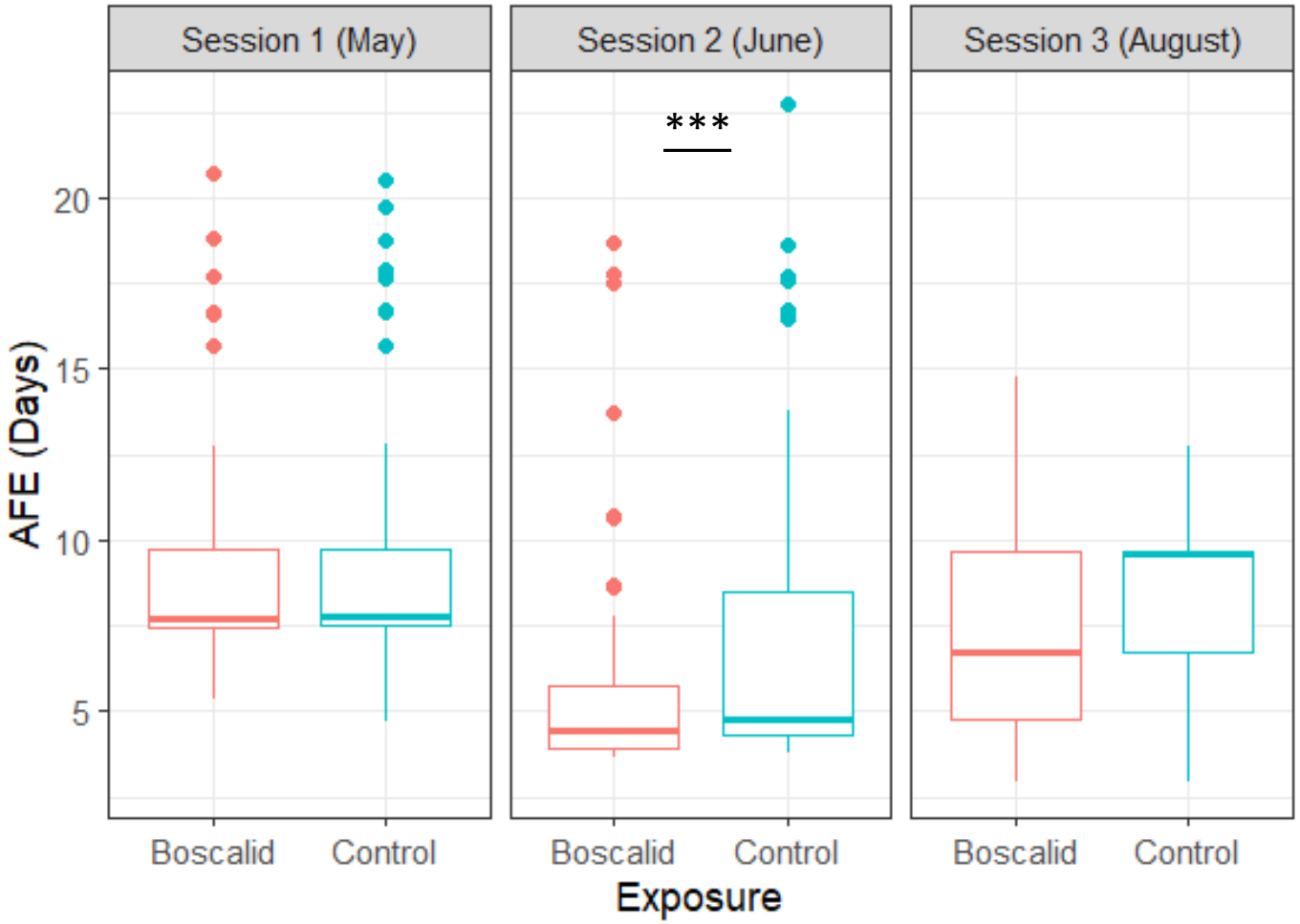


High effect between the different sessions

- Shorter life span of bees in replicates 2 and 1 compared
- No difference between modalities for replicate 1 and 3

- Significant difference between control and Boscalid modalities for replicate 2

Age of first exit (AFE) of *In vivo* bees

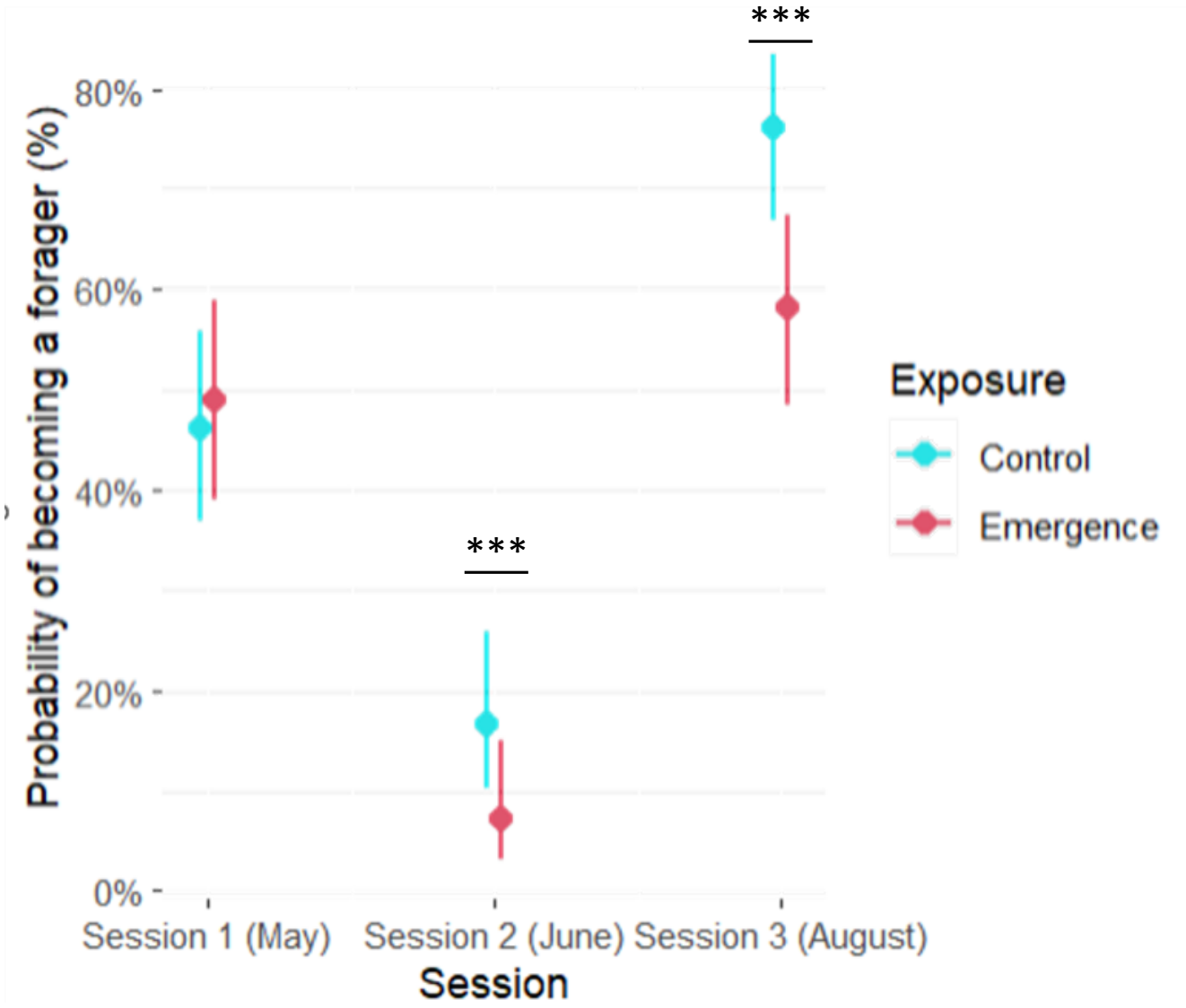


Hight effect between different sessions

- Session 1: bees leave out on average around 9.2 days at the sessions
- Session 2: -1.5 days
- Session 3: -1.2 days

- Significant effect of Boscalid on the age of first exit of bees during second session

Probability of becoming a forager and age at onset of foraging



Hight effect between different sessions

- Session 1: 50% around 9.2 days at the sessions
- Session 2: -1.5 days
- Session 3: -1.2 days

- Significant effect of Boscalid on the age of first exit of bees during second session