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Development of molecular tools to identify the main weevil pests of oilseed rape and their natural enemies.

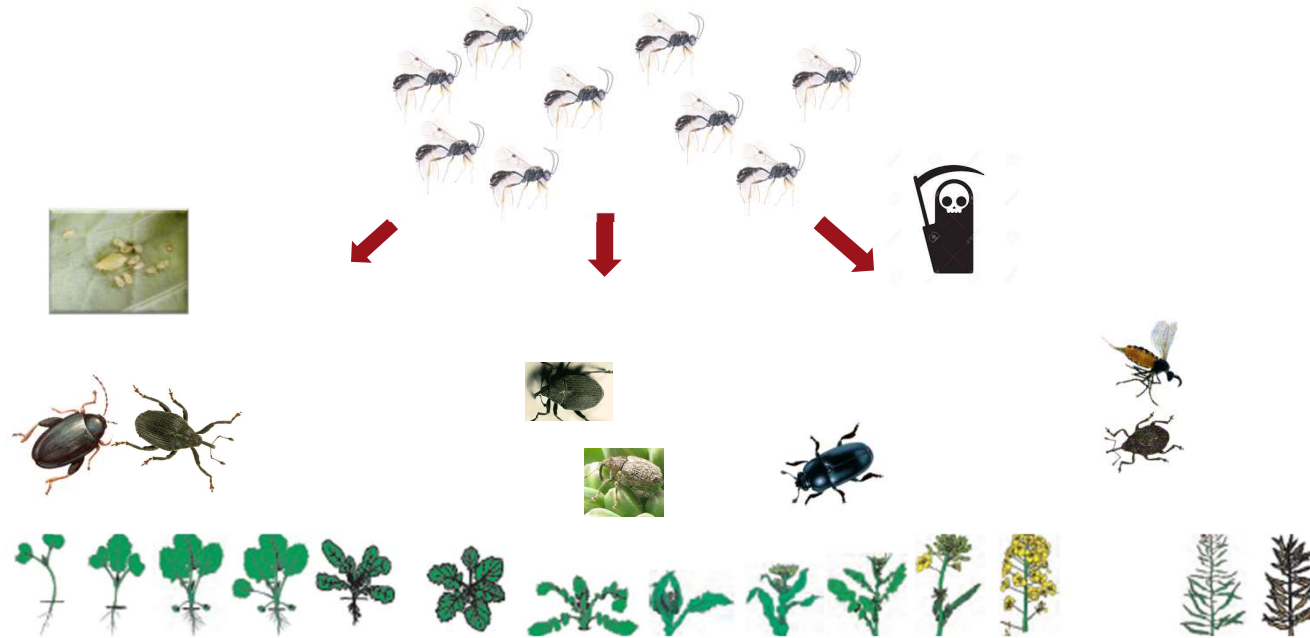
Robert C., Bothorel S., Luce S., Lauvernay A., Leflon M., Delvare G., Streito J.C., Pierre E., Cruaud P., Ollivier M., Genson G., Cruaud A., Rasplus J.Y.

Contact : c.robert@terresinovia.fr



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OSR pests regulated by many parasitoids



- Natural enemies can help farmers to regulate pests populations in their fields -> **conservation biological control**.
- To promote their action, a good knowledge of their biology is essential.

➡ **Tools to identify species and quantify parasitism rates are needed**
-> **Coleotool Project**





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A massive field collection for two years to create the database

Trapping



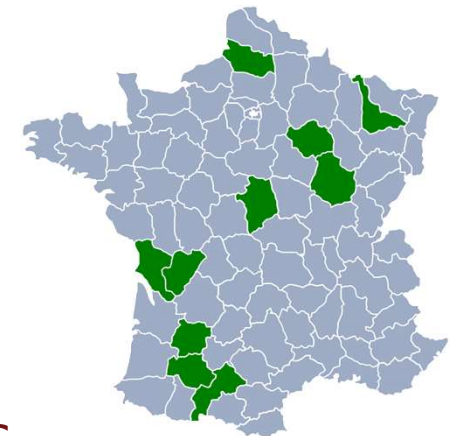
Rearing



159 collected samples in spring and in autumn 2014, in most French regions



35 larval populations of weevils were reared in 2014 and 2015 in most French regions



Sampling location





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Updated references on endoparasitism rates (PR)

C. picitarsis

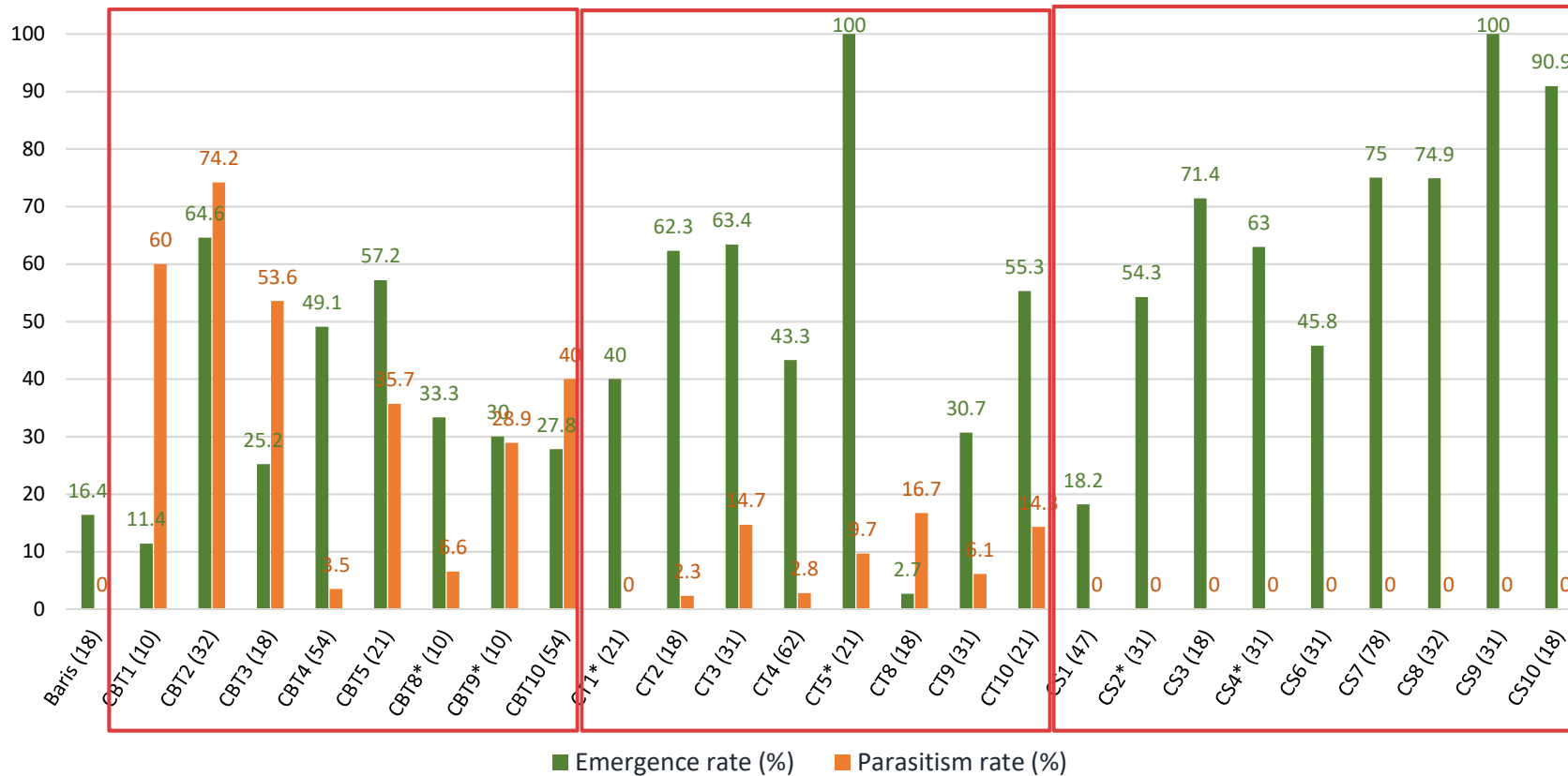
PR mean = 37.8 (sd=24.8)

C. napi or pallidactylus

PR mean = 8.3 (sd=6.4)

C. obstructus

PR mean = 0 (sd=0)





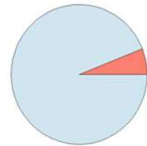
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Updated references on the parasitoids species and their territorial distribution



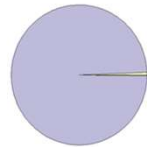
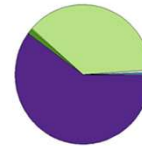
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Pests

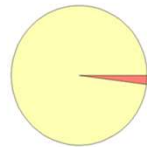
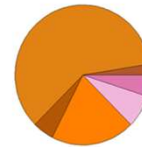


C. picitarsis
targeted

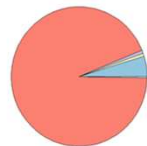
Parasitoids



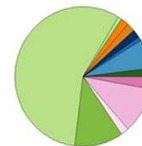
C. obstrictus
targeted



C. napi
targeted



C. pallidactylus
targeted



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Identified pests

- ☐ *Aulacobaris coerulescens*
- ☐ *Ceutorhynchus napi*
- ☐ *Ceutorhynchus obstrictus*
- ☐ *Ceutorhynchus pallidactylus*
- ☐ *Ceutorhynchus picitarsis*

Identified parasitoids

- ☐ *Anaphes* sp. 2
- ☐ *Aneucis melanaria*
- ☐ *Diospilus capito*
- ☐ *Diospilus oleraceus*
- ☐ *Diospilus* sp.
- ☐ *Mesopolobus morys*
- ☐ *Mesopolobus* sp.
- ☐ *Necremnus hippia*
- ☐ *Stenomalina gracilis*
- ☐ *Tersilochus fulvipes*
- ☐ *Tersilochus heterocerus*
- ☐ *Tersilochus obscurator*
- ☐ *Tersilochus*
- ☐ *Tersilochus stenocari*
- ☐ *Triaspis caudata*
- ☐ *Triaspis*
- ☐ *Trichomalus apertus*
- ☐ *Trichomalus perfectus*
- ☐ *Trichomalus* sp.

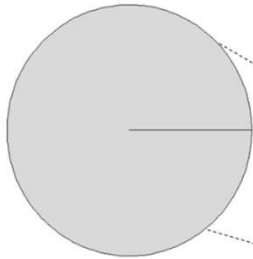


Updated references on the parasitoids species and their territorial distribution

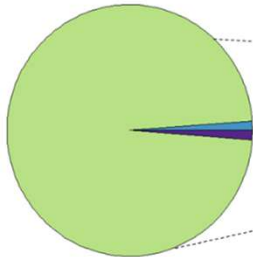
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Proportion of the different parasitoid species in samples targeting *C. picitarcis* (2014)

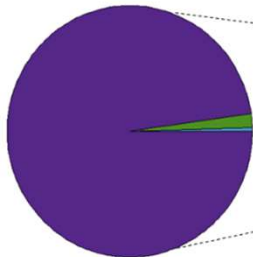
Number of parasitoids = 4



Number of parasitoids = 82
PR rate = 15%

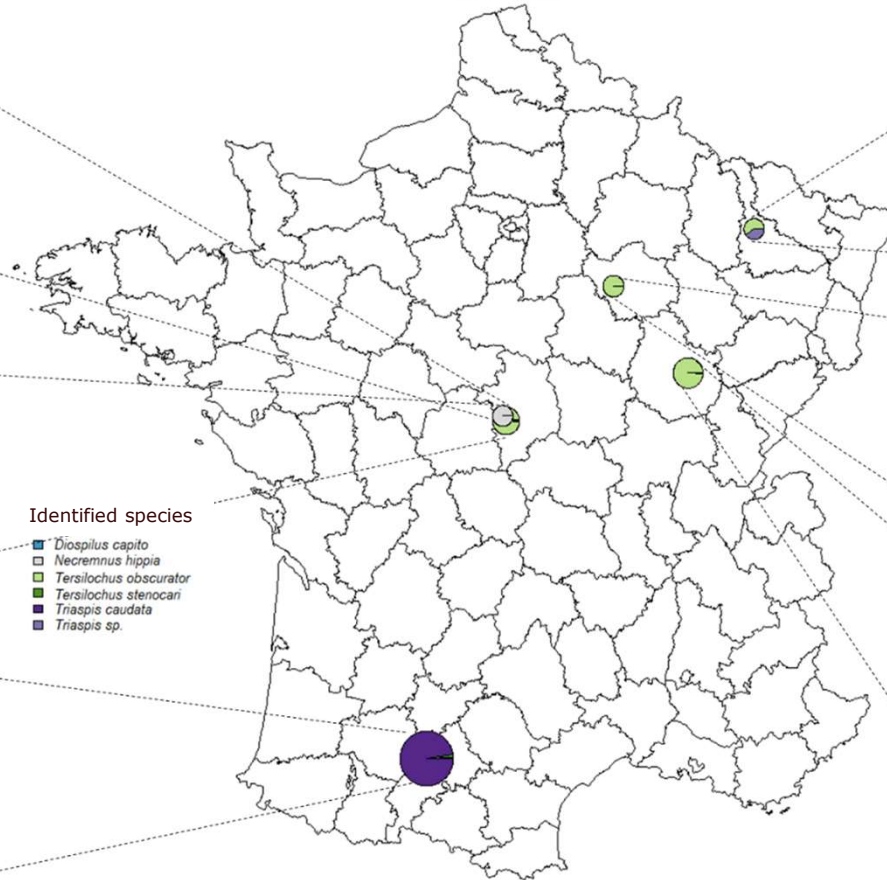


Number of parasitoids = 394
PR rate = 20%

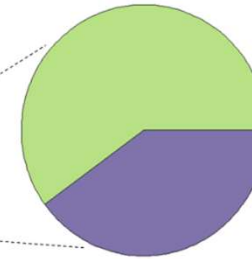


Identified species

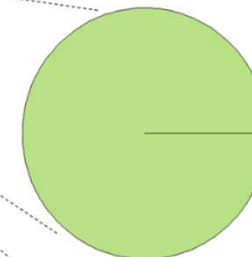
- *Diospilus capito*
- *Necremnus hippia*
- *Tersilochus obscurator*
- *Tersilochus stenocari*
- *Triaspis caudata*
- *Triaspis sp.*



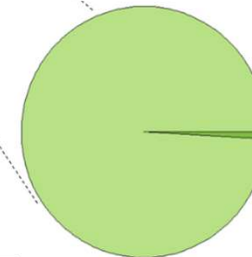
Number of parasitoids = 5
PR rate = 10%



Number of parasitoids = 12
PR rate = 19%



Number of parasitoids = 114
PR rate = 12%



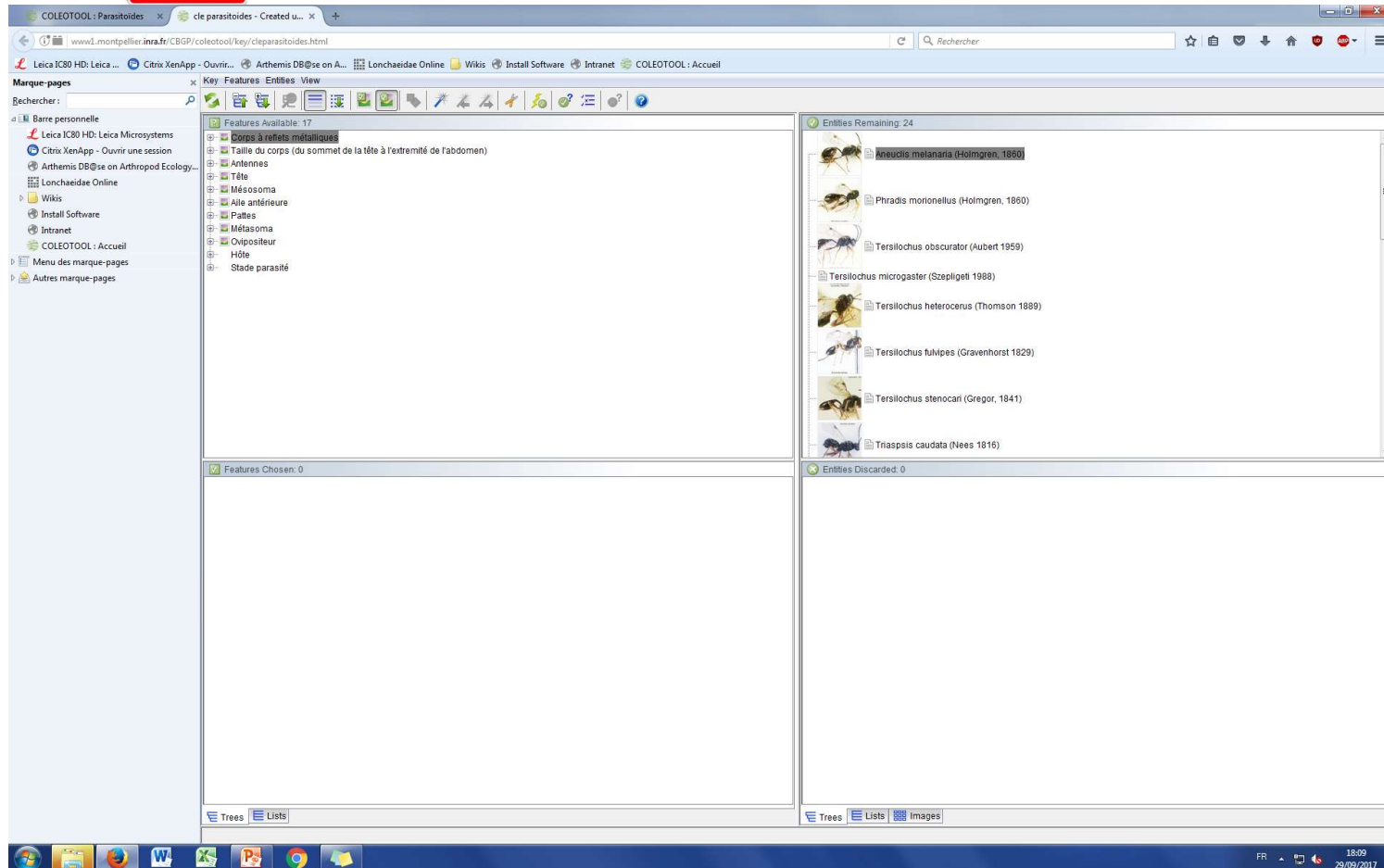


Creation of a morphological identification key for weevils (9 sp) and their parasitoids (23 sp)

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To identify parasitoids, the host must be known



<http://www1.montpellier.inra.fr/CBGP/coleotool/index.html>



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Creation of a morphological identification key for weevils and their parasitoids

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Creation of a morphological identification key for weevils and their parasitoids

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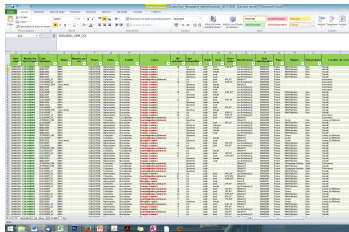
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Identification by entomologists

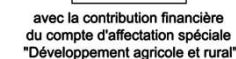
Specimen collection

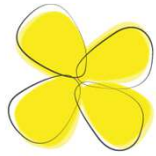


CO1 gene



**Database creation
(available online -> Arthemis
database)**





Creation of a database containing 149 validated sequences of weevils and their parasitoids

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Arthemis Home Search on Taxa Search on specimen(s) Identification tools Database information Contributors Sponsors Contact us Unknown user

Pairwise sequence alignment

☒ I have read [the disclaimer](#) and I agree with the conditions and limitations associated with the usage of the software

Pairwise sequence alignment parameters

Paste sequence to align:

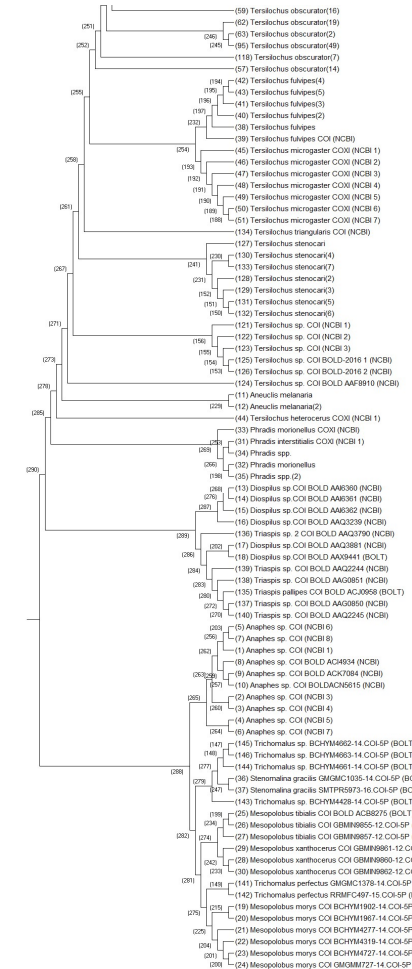
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qqagagggtgacccatttttataccagcatctatt
    
```

Start alignment

Draw Tree Expand Alignments Export data

Pairwise Results		Clustering results							
#	Reference description	Score	Probability	Similarity%	Fragments	Overlap%	Direction	Rating	
1	JSTR02652_0103 Ceutorhynchus napi, nlink892: COI Sequence	1044.49	0	100	1	100	+/+	*****	
2	EPIE00592_0102 Ceutorhynchus napi, nlink892: COI Sequence	1041.32	0	99.848	1	100	+/+	****	
	EPIE00592_0104								

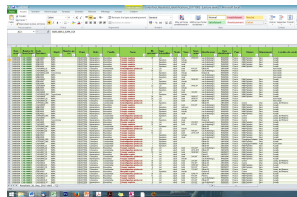
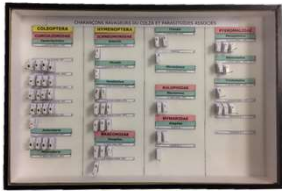




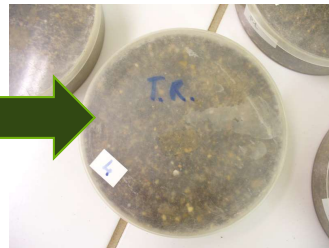
Development of molecular identification tools

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Specimen
collection

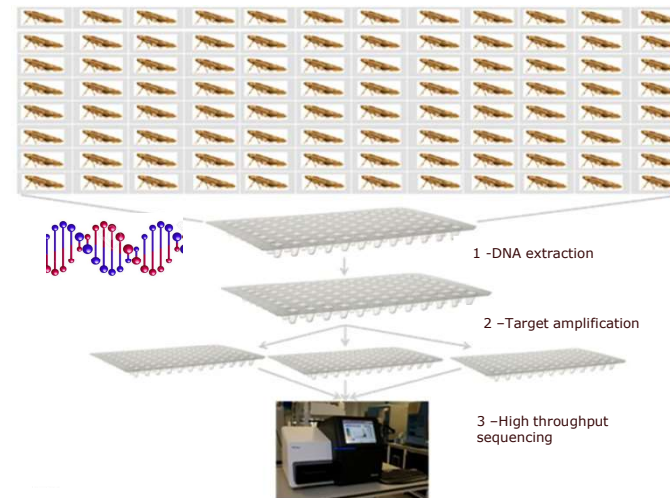


Rearing



Quantification of
parasitism rates
by rearing
parasitized
weevil larvae

High throughput sequencing
methods



- Allow the sequencing of several hundred insects in parallel
 - Cost reduction
- Allow to study trophic relationship

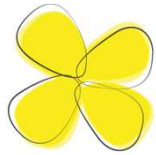




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Outlooks

- Development of simplified molecular methods to identify specimens at any stage and quantify parasitism rates on a regular basis (quick results, easy to use...)
- Development of tools for *Psylliodes chrysocephala* and their parasitoids.
- Practical application of the developed methods in the R2D2 project (poster)



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Tools available online on the Coleotool website

<http://www1.montpellier.inra.fr/CBGP/coleotool/index.html>

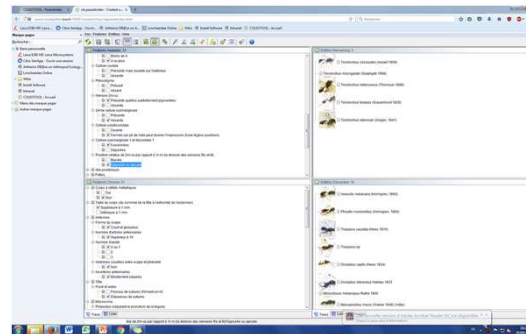
Sheets on species

Taxonomic information	
Complete taxon name	Teriochus (Teriochus) fulvipes (Gravenhorst, 1829)
Taxonomic rank	species
Classification	Animalia, Eumetazoa, Arthropoda, Hexapoda, Insecta, Hymenoptera, Apoidea, Ichneumonidae, Ichneumoninae, Teriochini, Teriochus (Teriochus)
Associated records	None
Taxon validity	nomen valid
Junior synonym(s)	1. Porizon fulvipes, 2. Teriochus gallicator, 3. Teriochus gallicator
Taxon iconography	

Link to:

<http://arthemisdb.supagro.inra.fr/>

Morphological identification key



Sequence database

Link to:

<http://arthemisdb.supagro.inra.fr/>

Thank you for your attention !



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