



Insect chemosensory receptors: contribution to plant-insect relationship

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SCIENCE & IMPACT

Sensory ecology department

Insect chemical ecology, from signal identification to their receptor and behavior characterization

Crop pests



Parasitoids



Disease vectors



Chemical senses of insects



Credit: web

Objectives of our group

- ✓ How the insect selects the good chemical information in a complex environment?

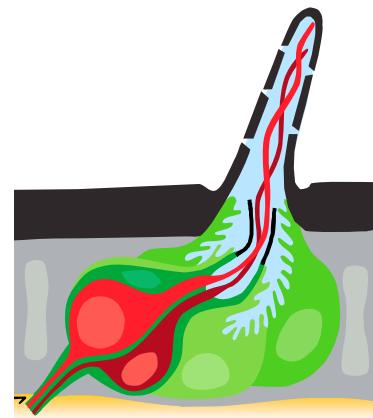
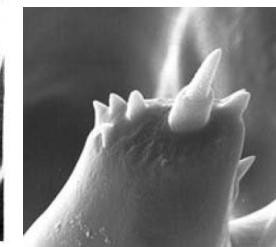
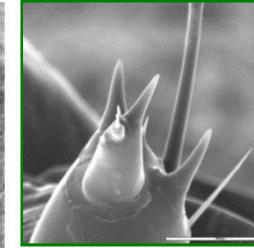
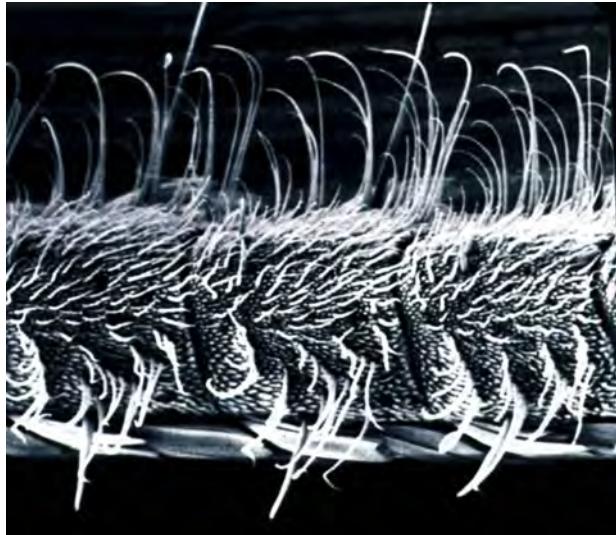
Understanding the **peripheral** mechanisms
Special focus on chemosensory receptors

- ✓ Do and how the chemosensory receptors shape the animal sensory ecology

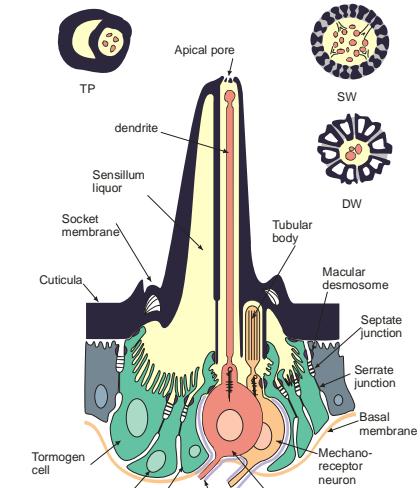
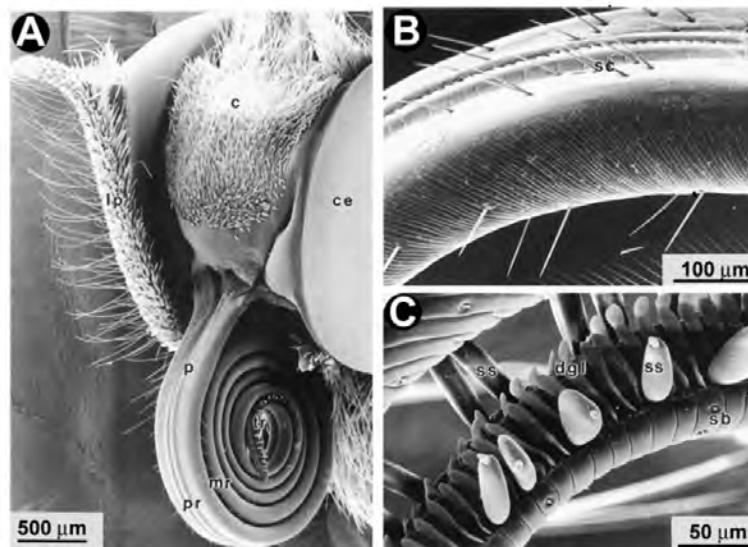
Evolution of gene families

- ✓ Identification of receptors as new targets for pest controls

A morpho-functional unit: the chemosensory sensilla

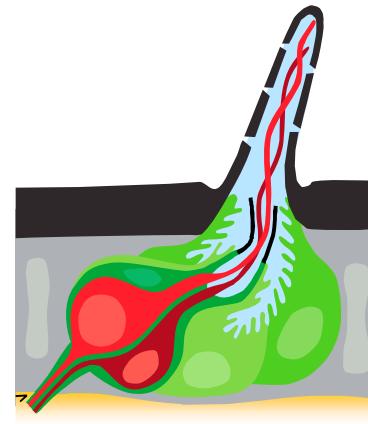


Olfactory sensilla

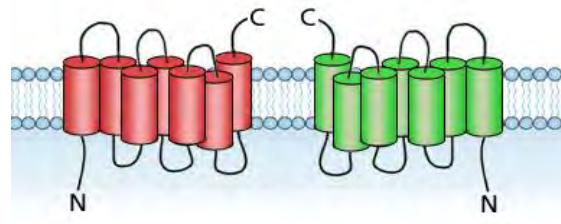


Tatse sensilla

Insect chemosensory receptor families



ORs

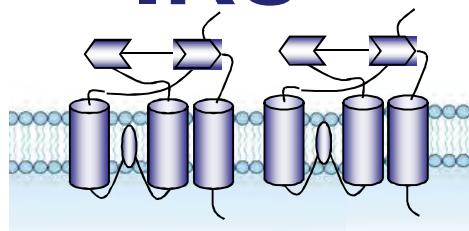


odorant
receptor

Co-receptor
ORco

Odors
pheromones

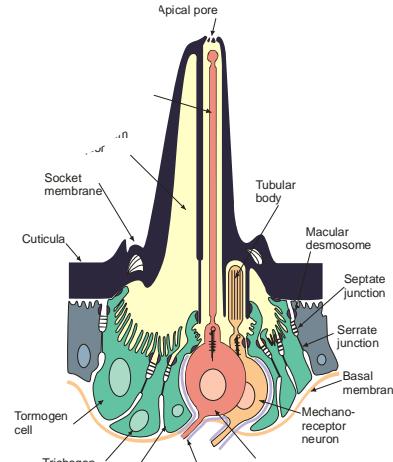
IRs



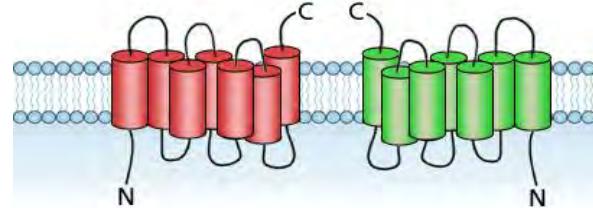
Co-receptor
IR8a/25a

ionotropic
receptor

Fermentation products
acids



GRs



Gustatory
receptor

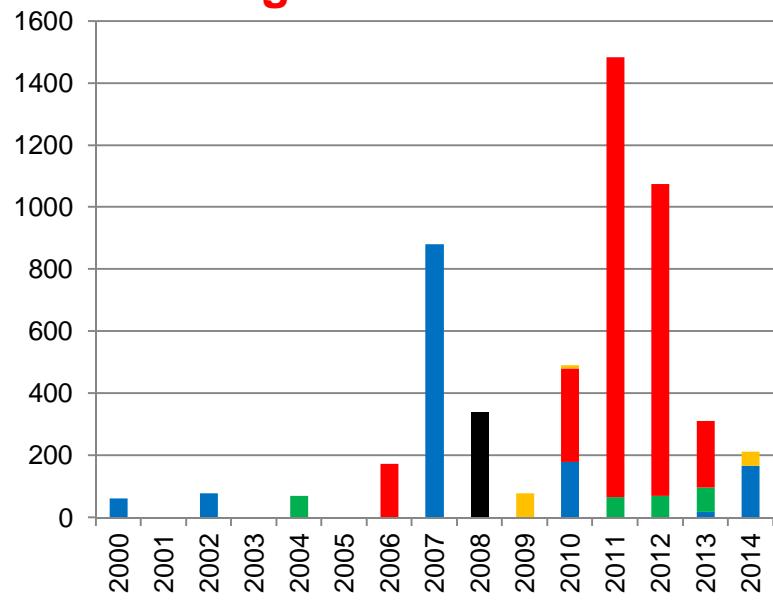
Co-receptor?

CO₂, Sugar
Bitter compounds
pheromones

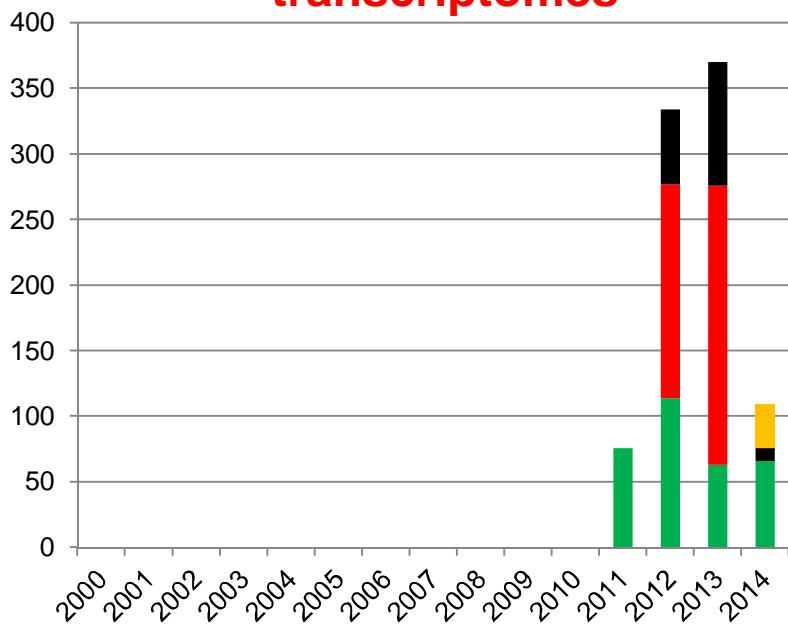
Development of high-throughput sequencing methodologies: An increasing number of chemosensory gene repertoires from a wide diversity of insect species



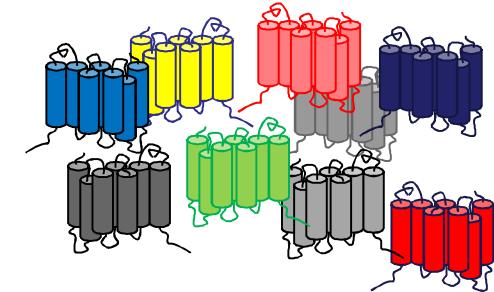
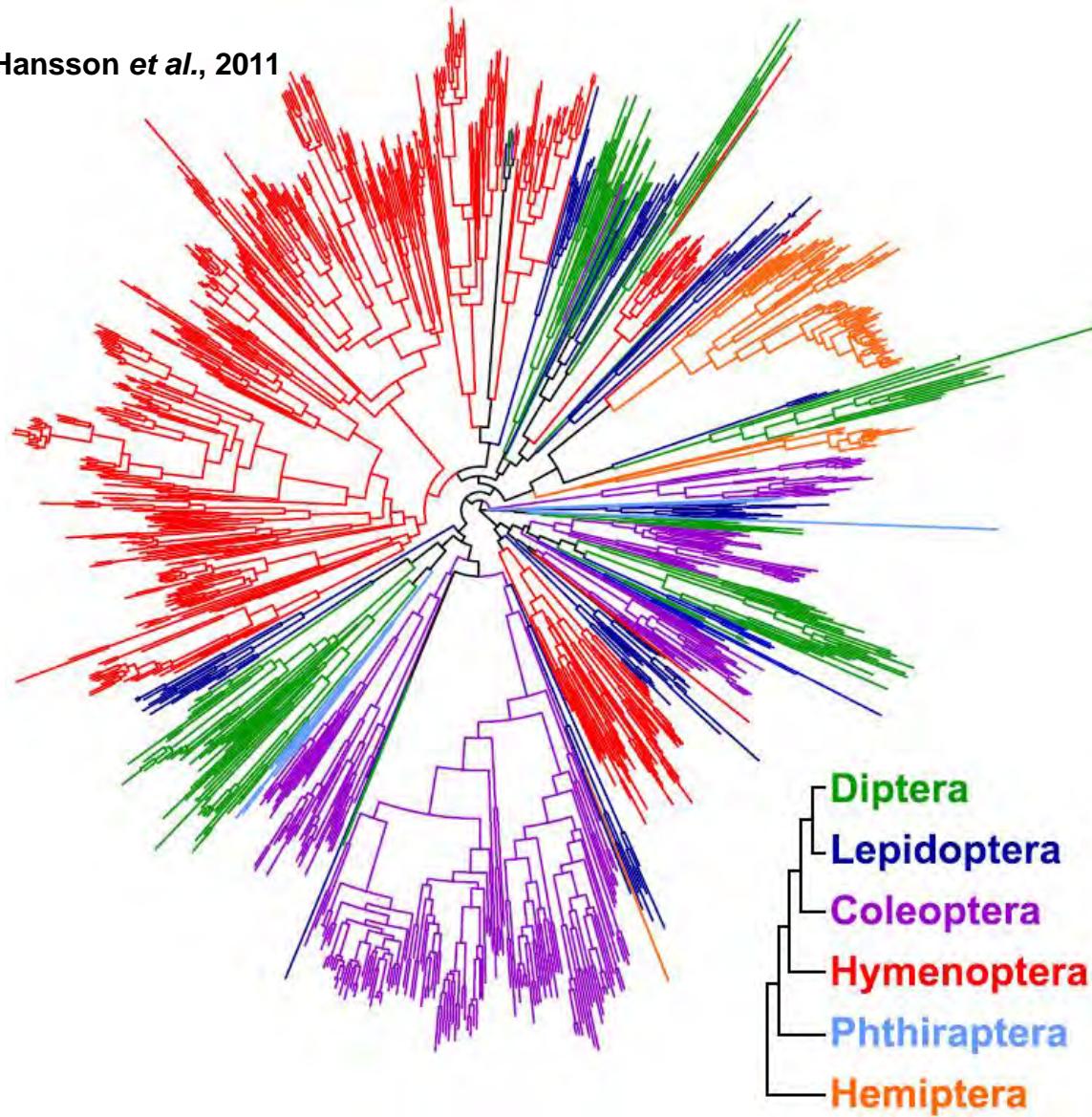
Evolution of the number of insect olfactory receptors identified through analysis of
genomes **transcriptomes**



- Others
- Coleoptera
- Hymenoptera
- Lepidoptera
- Diptera



Hansson et al., 2011



- 10-400 ORs/species
- Rapidly evolving
- Very divergent between and within species
- But most ORs are still orphan receptors

How these ORs evolved to adapt to the species ecology?
How a species use its OR repertoire to deal with its environment?

Large OR repertoires have been functionally characterized in only two model species



*Drosophila
melanogaster*



*Anopheles
gambiae*



Although diverse, Diptera can not resume insect chemical ecology

The cotton leafworm *Spodoptera littoralis* (*Noctuidae*) a model species for herbivores

- highly polyphagous crop pest
- a strong background on olfactory behaviors and physiological properties of olfactory neurons
- a long list of molecules known to be active on either behavior and/or antennae
- 48 Ors identified via transcriptomics



Legeai et al 2011, BMC G
Jacquin-Joly et al 2012, IJBS
Poivet et al 2013, PloS One



The current challenge: development of functional genomics in non model insects



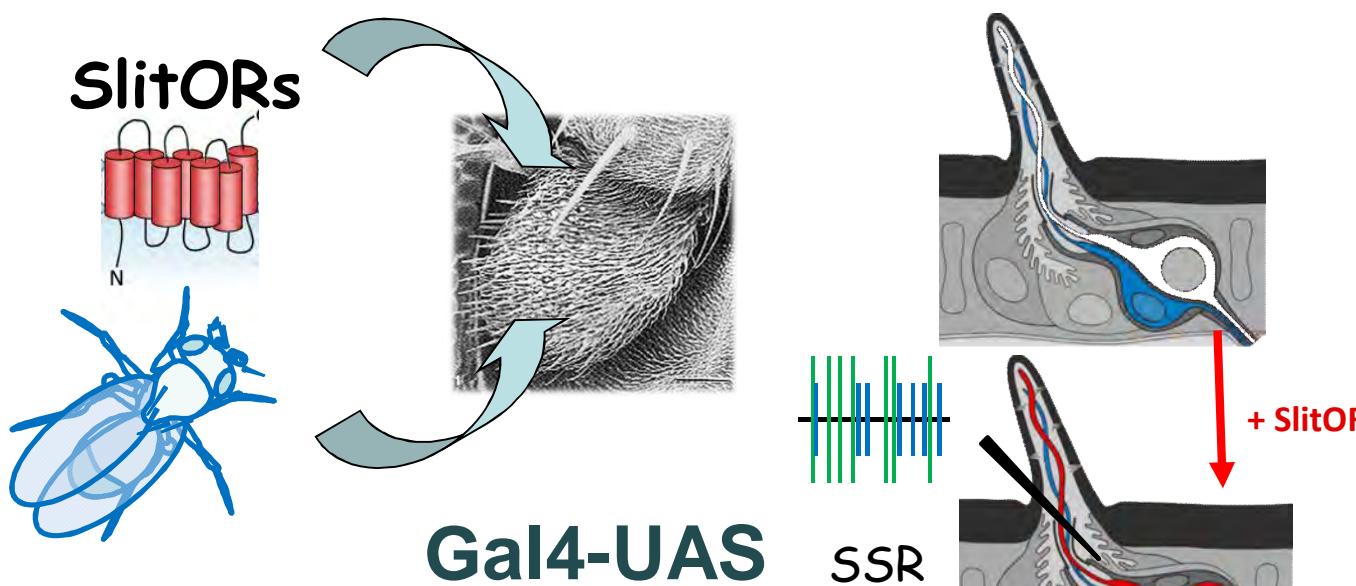
*Heterologous expression:
deorphanization of ORs*



*Reverse genetic:
development of genome editing*

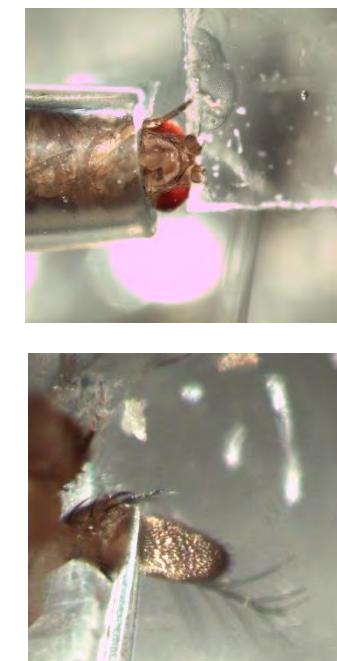
OR deorphanization using the *Drosophila* « empty neuron »

iEES Paris



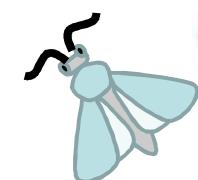
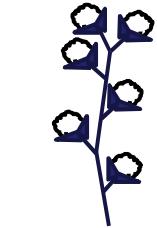
Montagné et al 2012 *Eur J Neurosci*
De Fouchier et al 2015 *Front Ecol Evol*
De Fouchier et al submitted

Hallem et al 2004 bs
Kurtovick et al 2007 ts



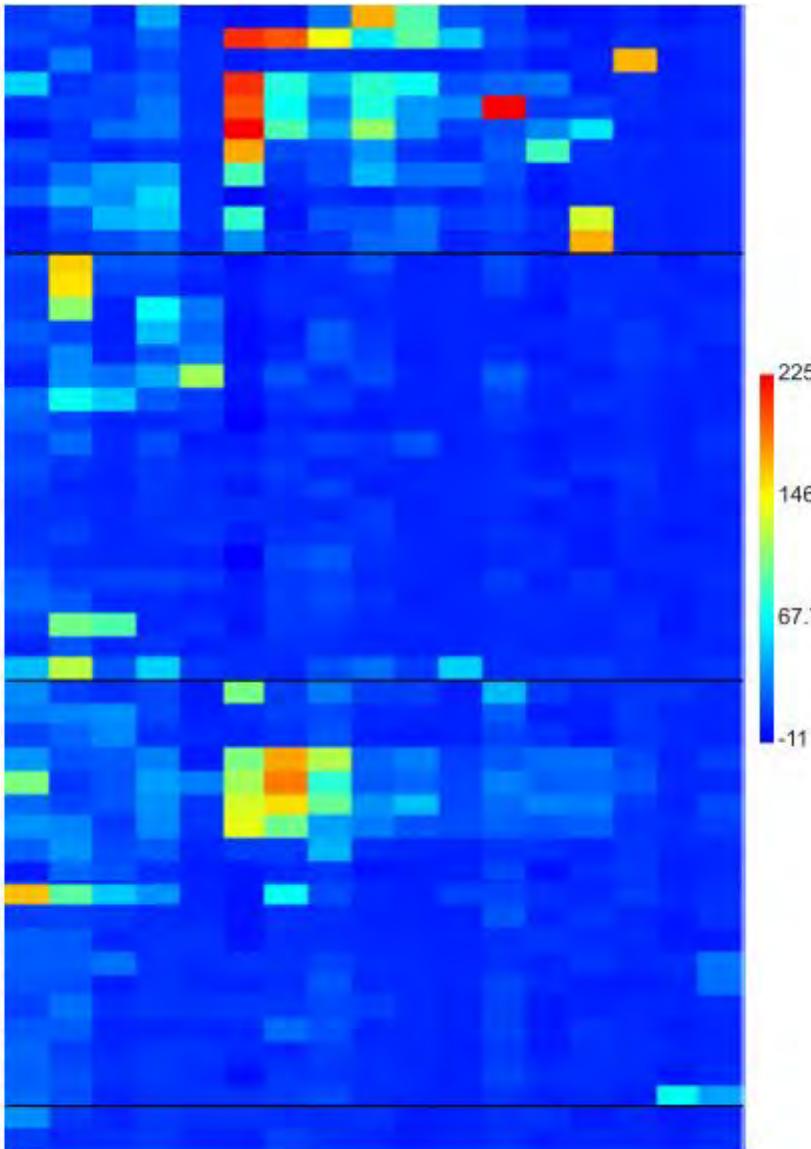


A. De Fouchier



indole
benzyl alcohol
eugenol
benzaldehyde
2-phenylacetaldehyde
acetophenone
1-indanone
benzyl methyl ether
estragole
methyl benzoate
methyl salicylate
(E)-ocimene
 β -myrcene
3-carene
 α -pinene
 β -pinene
 (\pm) -linalool
geranol
thymol
carvacrol
 α -humulene
 β -caryophyllene
 α -opaene
(E,E)- α -farnesene
(E,E)-farnesol
 (\pm) -nerolidol
 (\pm) -phytol
DMNT
TMTT
sulcatone
*(E)*2-hexenal
nonanal
decanal
*(E)*2-hexenol
*(Z)*3-hexenol
1-hexanol
1-heptanol
1-octanol
1-nonal
*(Z)*3-hexenyl acetate
EDD
14:OAc
*(Z)*7-12:OAc
*(Z)*9-14:OAc
*(Z)*9-14:OH
*(Z)*11-14:OAc
*(E)*11-14:OAc
(Z,E)-9,11-14:OAc
(Z,E)-9,12-14:OAc
(Z)-jasmine
methyl jasmonate

SlitORs



One OR can recognize several odorants

One odorant can be recognized by several ORs

development of targeted mutagenesis

- Loss of-function studies using RNAi are generally difficult to achieve in Lepidoptera
Terenius et al. 2011
Kobayashi et al. 2012
Shukla et al 2016
- Transgenesis has been for long restricted to the silkworm moth model and with random events...
Tamura et al 2000

...Until the development of new genome editing methods

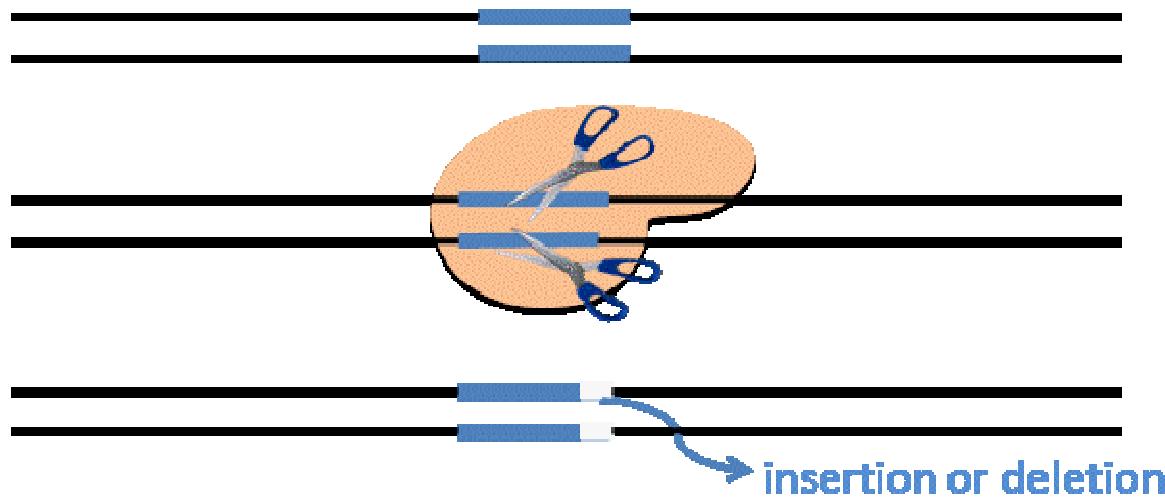
Photo: M. Renou, iEES

Genome engineering by targetable nucleases

- ➡ ZFN
- ➡ TALEN
- ➡ CRISPR/Cas9

Zinc finger nuclease
Transcription activator-like effector nuclease
Clustered Regularly Interspaced Short Palindromic Repeats

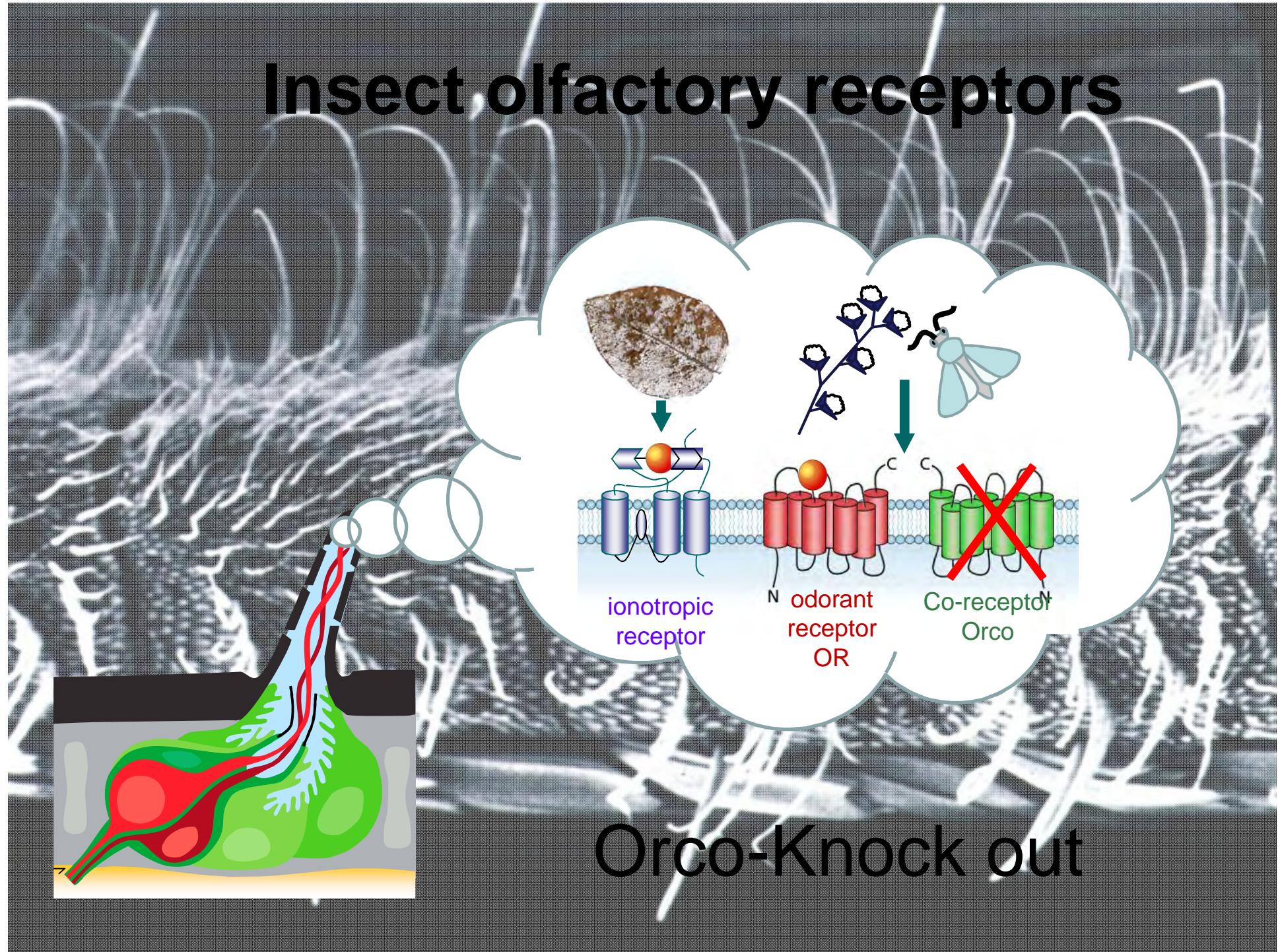
induce specific breaks in DNA whose repair either induces local mutations or stimulates homologous recombination



A challenge in non-model Lepidoptera

But recent developments

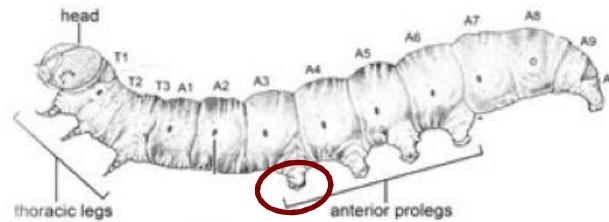
Insect olfactory receptors





F. Koutroumpa C. Monsempes MC. François

GO Genotyping -results



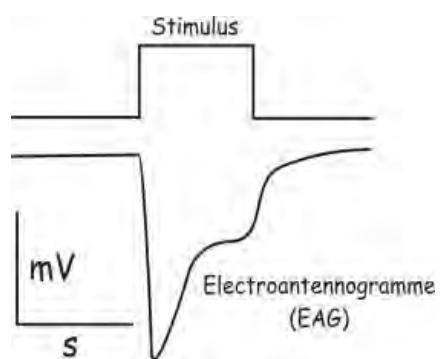
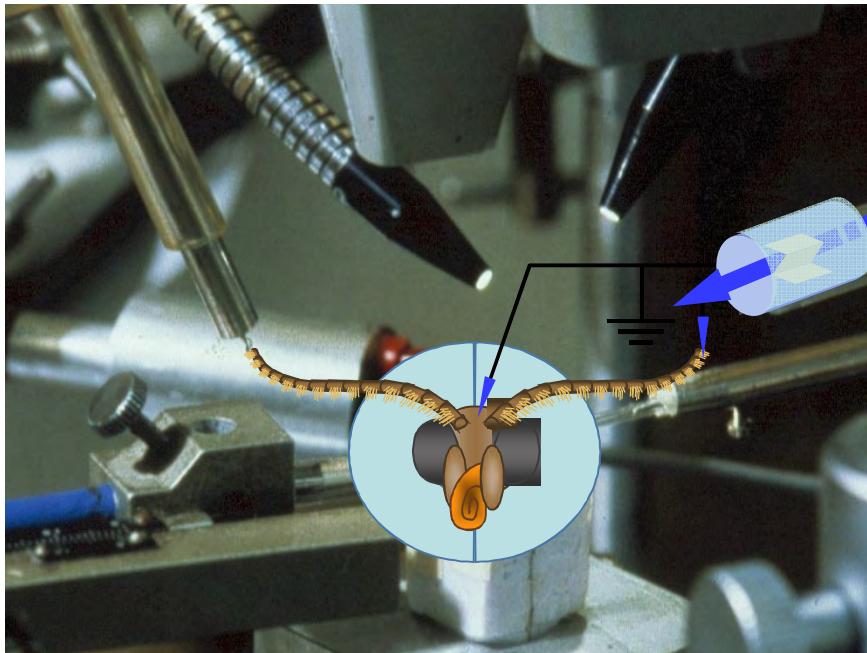
F1-29.1.1...	GCGATTCTCATCACCATACATTTGCCTGTAAGGAACGTTAAAAAAATTACACAGAAAATTAGTATGGTATGGTAGGTAGCACAC	56+
F1-30.3.11	GCGATTCTCATCACCATACATTTGCCT GTAATACAGGTATCATACATTCTGCCA GGGTATCAACATGGCC CAGTACTCCGATG	24
F1-31.3.6	GCGATTCTCATCACCATACATTTGCCT GTAT SRRYATSTTT GGGTATCAACATGGCC CAGTACTCCGATGAGGTCAACGAGCT	10
F1-29.1.2	GCGATTCTCATCACCATACATTTGCCT GTAT SCTGCCT GGGTATCAACCTGGCC CAGTACTCCGATGAGGTCAACGAGCTGAC	7
F1-30.3.4..	GCGATTCTCATCACCATACATTTGCCT GTACTCCGAT GGGTATCAACATGGCC CAGTACTCCGATGAGGTCAACGAGCTGACT	7
F1-29.1.13	GCGATTCTCATCACCATACATTTGCCT GTATGGGTAT SRGTMW CAACATGGCC CAGTACTCCGATGAGGTCAACGAGCTGACT	6
F1-31.1.9	GCGATTCTCATCACCATACATTTGCCT GTGT GGGTATCAACATGGCC CAGTACTCCGATGAGGTCAACGAGCTGACTGCCAA	3
F1-31.1.6	GCGATTCTCATCACCATACATTTGCCT GTT : GGGTATCAACATGGCC CAGTACTCCGATGAGGTCAACGAGCTGACTGCCAAC	2
F1-30.1.1	GCGATTCTCATCACCATACATTTGCCT GT : GGGTATCAACATGGCC CAGTACTCCGATGAGGTCAACGAGCTGACTGCCAAC	2
F1-31.1.2	GCGATTCTCATCACCATACATTTGCCT ::: GGGTATCAACATGGCC CAGTACTCCGATGAGGTCAACGAGCTGACTGCCAAC	4
F1-29.1.14	GCGATTCTCATCACCATACATTTGCCT ::::: GTATCAACATGGCC CAGTACTCCGATGAGGTCAACGAGCTGACTGCCAAC	6
WT	GCGATTCTCATCACCATACATTTGCCT GTATGGGTATCAACATGGCC CAGTACTCCGATGAGGTCAACGAGCTGACTGCCAAC	WT

PAM

gRNA

**13 different mutations observed,
most with interrupt coding sequence**

G2 Phenotyping electroantennography (EAG)



Odorants tested:

E-ocimene

Z3-hexenyl-Ac

E2-hexenol

benzyl OH

PAA

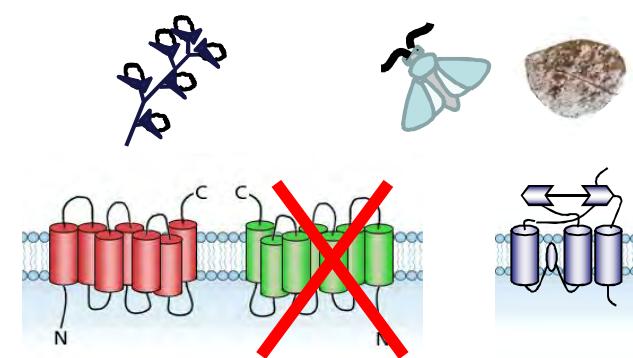
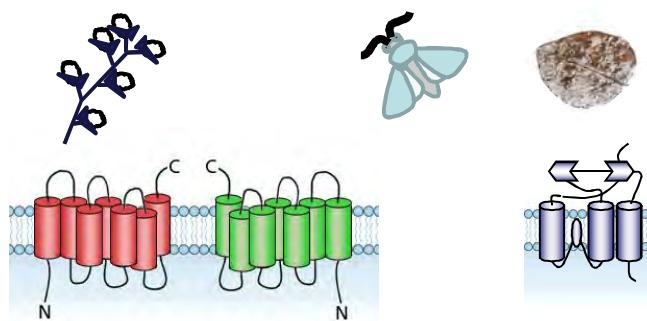
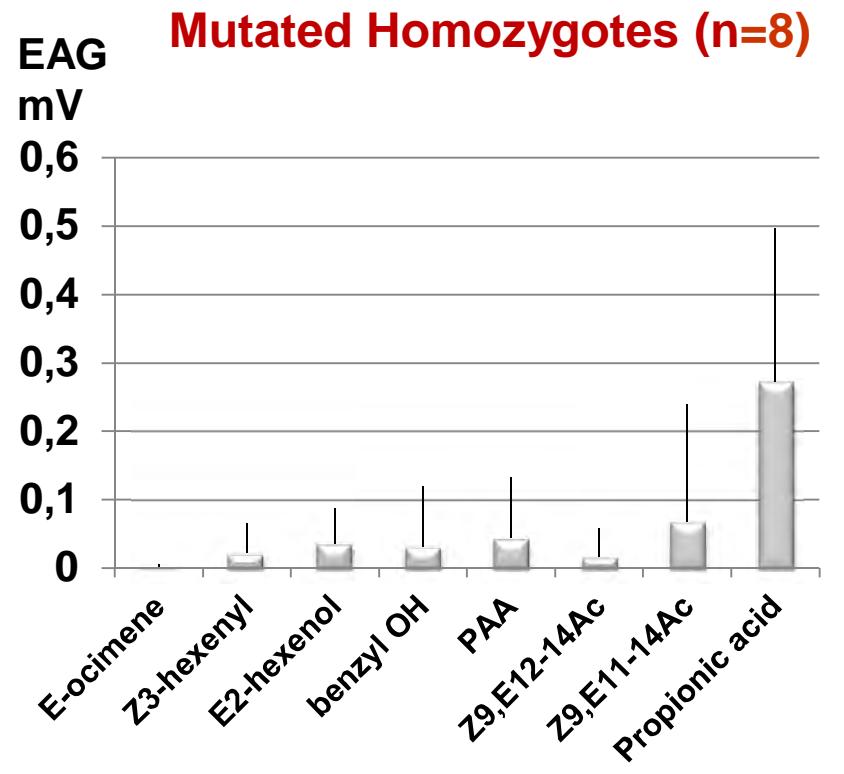
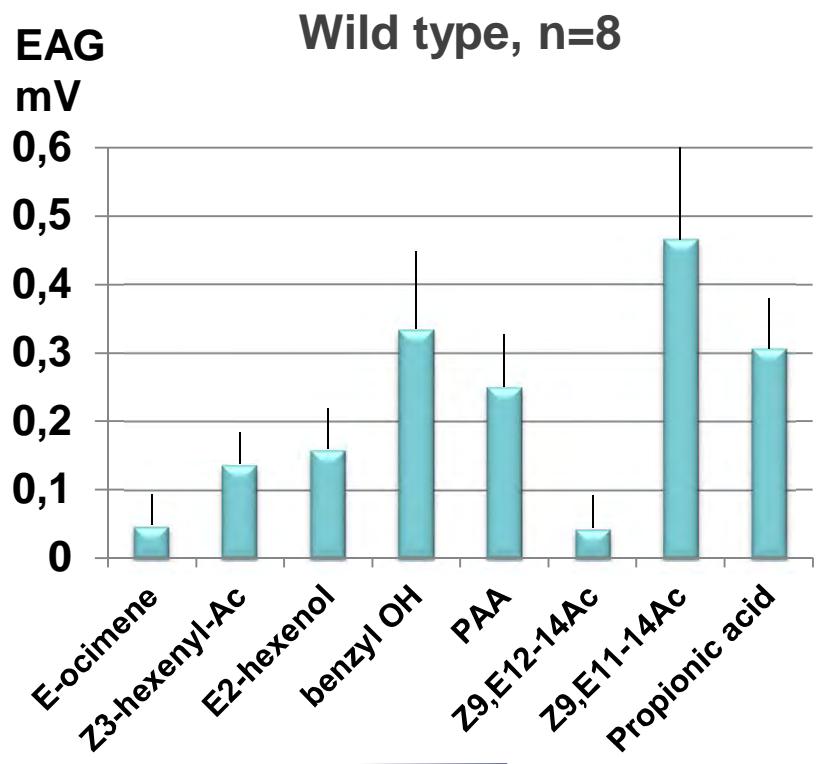
Z9, E12-14:Ac

Z9, E11-14:Ac



IR

Phenotyping G2



Conclusion

still far to understand how insects evolved different chemoreceptors to ensure species isolation and to satisfy their ecological needs

a need of more sequence data and functional data in a diversity of species from different taxa and with diverse ecology

NGS: affordable transcriptomics and genomics

Development and optimization of functional genomics:

Development of high-throughput screening approaches

Enlargement of the ligand spectra

CRISPR/Cas9 as a promising tool for non model species

ANR

DEMETER
2017-2021



evidences of functional grouping of ORs:



insight into their functional evolution

identified targets in pest control

Neglected IRs and GRs!

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Arthur de Fouchier, PhD

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Aurore Gallot, post-doc

Olivier Mirabeau, post-doc

Fotini Koutroumpa, post-doc

Marie-Christine François

Christelle Monsempes





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**Jean-Paul Concorde, Anne De Cian
Corrine Royer**

