Sex perfumes against agricultural insect pests: perspectives and challenges

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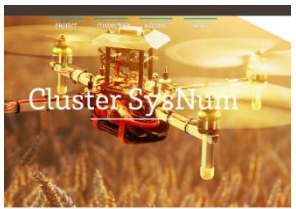

Workshop 22-23 nov 2018

ABSTRACT

Mating disruption (MD) is a powerful and sustainable control strategy against different types of mainly Lepidoptera agricultural pests. Since the first large scale trials on oriental fruit moth in Australia (1975), it is used for now more than 30 years against several pest insects, with very good efficiency against tortricid pests (orchards and grapes). This technique is growing up in french viticulture, with ca. 80,000 ha in 2018 (increase 3 times fold in 7 years). This technique of control is based on mating inhibition and is the result of different physiological and fine behavioral mechanisms (e.g zigzagging flight, counterturning when leaving the odor plume, etc…). It is a valuable and promising technique in the reduction of insecticide use. This semiochemical (pheromone) based technique targets the sexual behavior of pests by inhibiting sex partners meeting and thus mating. It is thus pheromone concentration dependent in the air , and most of the time the best results are obtained with a high and stable concentration cloud above the crop. Reversely, failures in efficiency often relies on bad or uncontrolled diffusion of the pheromone in the air. Here we present the basis of MD and our Sys Num research project on designing pheromone sensor for improving the pheromone diffusion and thus mating disruption against the fruit tortricid moths. (see the following talk by Petra Ivaskovic). This talk is presented within the Lab. Cluster 'SysNum'. Our project objective is to improve data collect in the environment via sensors and mathematical spatial modeling of the pheromonal 'cloud' above the crop.

Updated 27th Nov 2018

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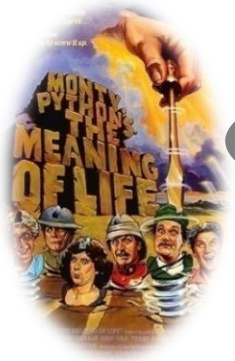


Agricultural pests are animals with reproductive strategies: be successful in different tasks

Drink (to live longer)

Mate

Oviposit

Wait safely when adverse conditions

Move (sometimes escape)

Live hidden (rest of the time)

Eat

Shelter

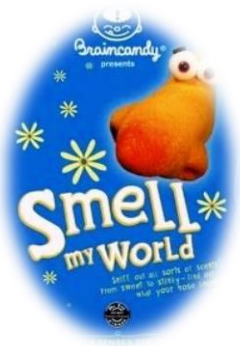
Escape or defend against adversity

Become an adult

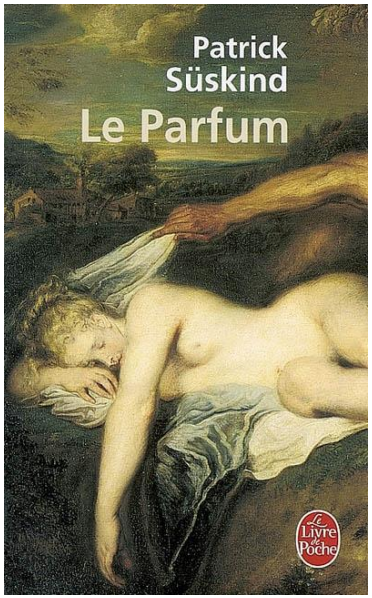
Mating… a critical step in life (?)

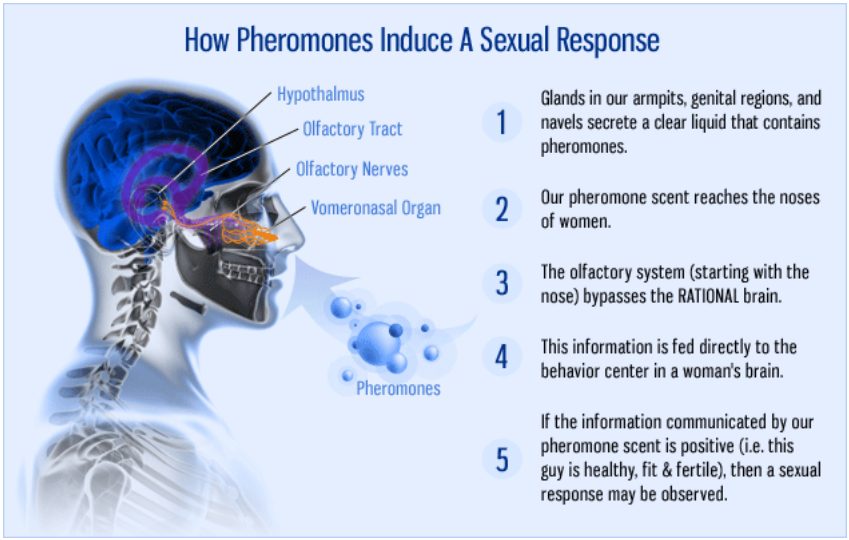
Efficiency to quickly mate is especially critical in small short lived insects with limited displacment.

This necessarily relies on trustable information and this will condition population size



Live in an odorant world





How to remain indifferent to a such beautiful partner ?

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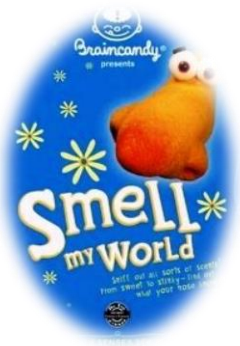
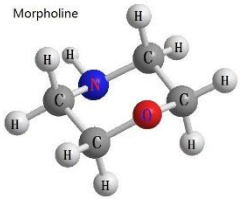


Live in an odorant world

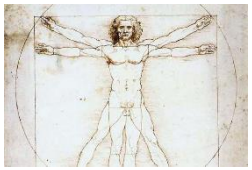
A pheromone is a chemical messenger, emited outside an organism which mediates inter-individual interactions with conspecific.

e.g.: mother kids, sex partners, alarm recruitment, trail following, colony structure, etc…

In fish too: e.g.morpholine in salmonids



Invertebrates/Vertebrates



Biol. Rev. (1984), **59**, *pp.* 333–388
Printed in Great Britain

HOMING AND OLFACTION IN SALMONIDS:
A CRITICAL REVIEW WITH SPECIAL REFERENCE TO
THE ATLANTIC SALMON

By OLE B. STABELL

Sex pheromones in bacteria

from J. R. Saunders

THE role of sex pheromones in mating is well established in higher organisms and fungi. In contrast there is scant evidence for such chemical sex attractants in bacteria. In Gram-negative bacteria the formation of aggregates of bacteria during conjugation is well known (see for example, Achtman & Skurray in *Microbial Interactions* (Series B, Receptors and Recognition Vol 2) 233 (ed. Reissig) Chapman and Hall, London, 1977). The molecular mechanisms involved in genetic transfer during bacterial conjugation have been intensively studied but much less is known of the signalling processes which might initiate contact

Nature volume 275: 692–694 (26 October 1978)
Bacteria communicate using peptides

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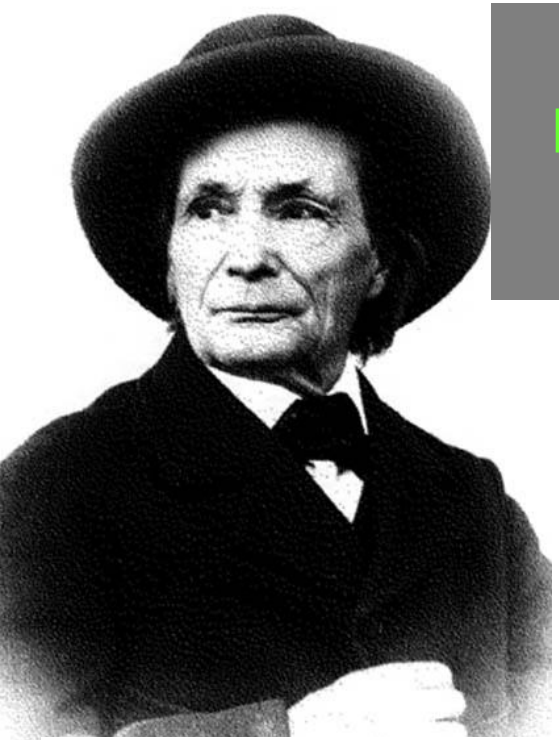
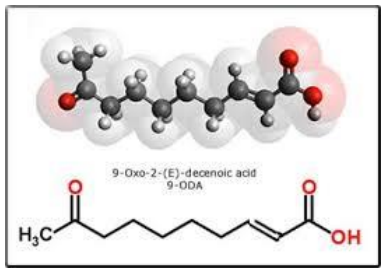
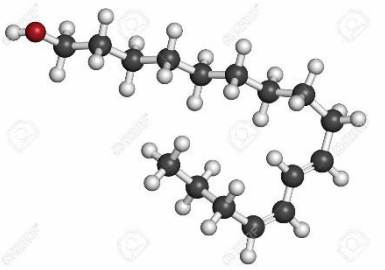


Chemical communication: Fabre was a pioneer… Von Frisch a nobel prize

Fisrt chemical identification of a sex peheromone in insects: Honeybee queen, 1958 Barbier (CNRS)
(…but not published fast enough).



Then Bombykol (sex pheromone *Bombyx mori* –vers à soie), 1959 (Butenandt).



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How works sex attraction in males Lepidoptera?

Females bears a 'phero' gland, and for nocturnal species (moths) emit at dusk few (1 >>> hundred) nanogrammes of a pheromonal cocktail that attracts conspecific males.

Reaching a plume of pheromone males initiate an upwind flight over hundreds of m up to the punctual source (female or bait), in typical zig-zagging. They adapt their flight speed to the wind speed.

Another characteristic behavior is 'counteturning' which allows staying in the plume. At the plume boundary (when odor concentration decreases), as soon as perceive a strong reduction in odor concentration,males calculate and memorize their angle from the wind axis and rapidely turn a complementary angle,

	Constituant majoritaire	Nom chimique
Lobesia		E7, Z9 dodecadienyl acetate
Eupoecilia		Z 9 dodecenyl acetate
Sparganothis		E 9 dodecenyl acetate
Argyrotaenia		Z 11 tetradecenyl acetate

Beautiful picture made by Stephan Rauscher: 'calling' female Lobesia botrana extruding its phero gland

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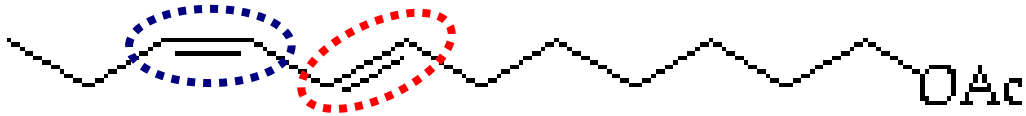
Lepidopteran moths produce a ‘phero’ cocktail

- Mostly linear C12-C14 hydrocarbons more or less oxygenated
- One very major constituent, and others at low concentration
- Each constituant has a function



E7,Z9-12Ac = (E,Z)-7,9-dodecadienyl acetate

E7 Z9 12 Ac	1
Z7 Z9-12 Ac	0.02
E7 E9-12Ac	0.01
Z7 E9-12Ac	0.01
E7 Z9-12Oh	0.25
Z9-12Ac	0.1
E9-12Ac	0.005

+ autres composés minoritaires

Arn et al. 1988, Agric. Ecosystems & environment

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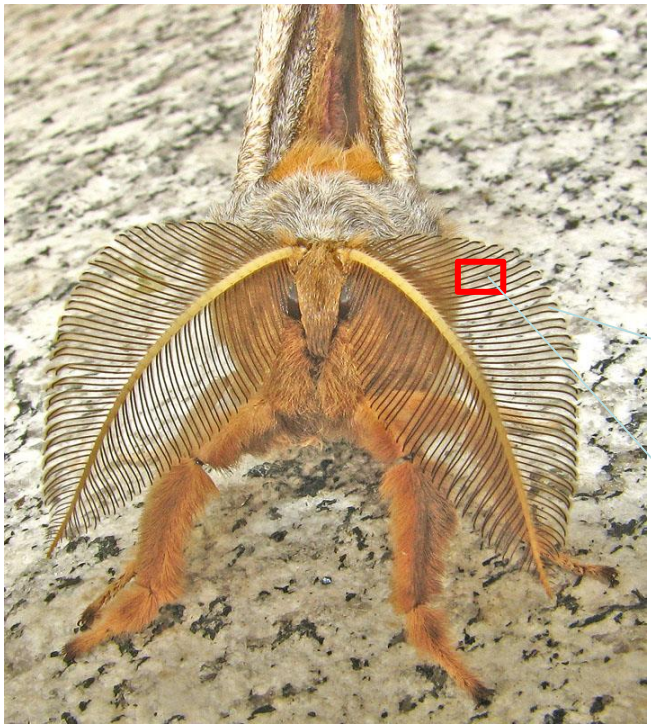
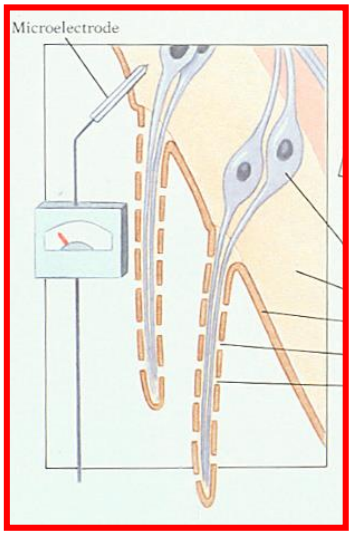
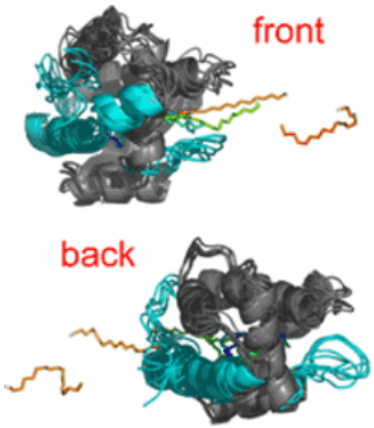
Back to the beautiful antennae of male *Bombyx mori*…

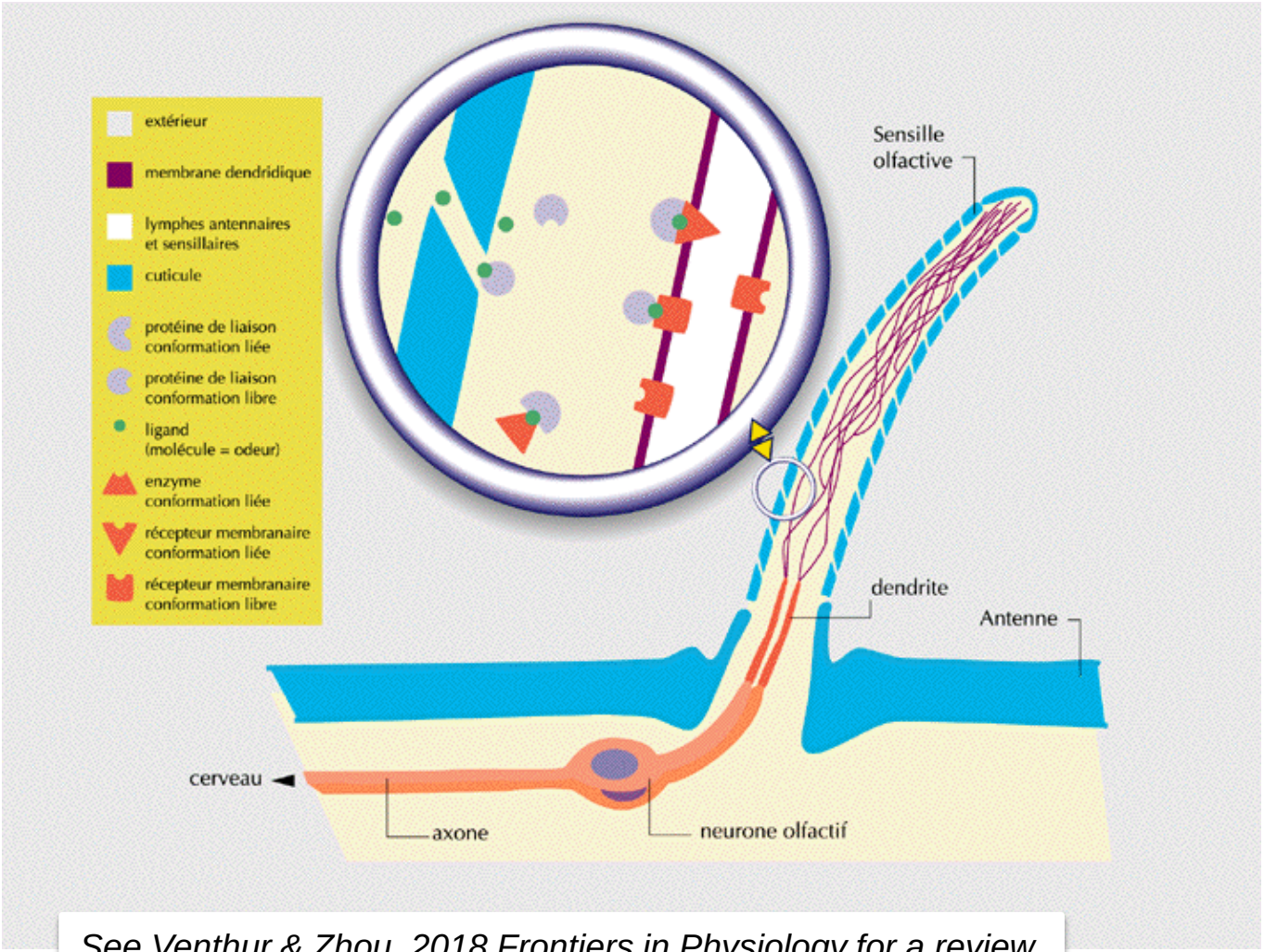
Antenna bears the olfactory receptor cells





Odour perception: two antennae one brain, detection and integration





See Venthur & Zhou, 2018 Frontiers in Physiology for a review

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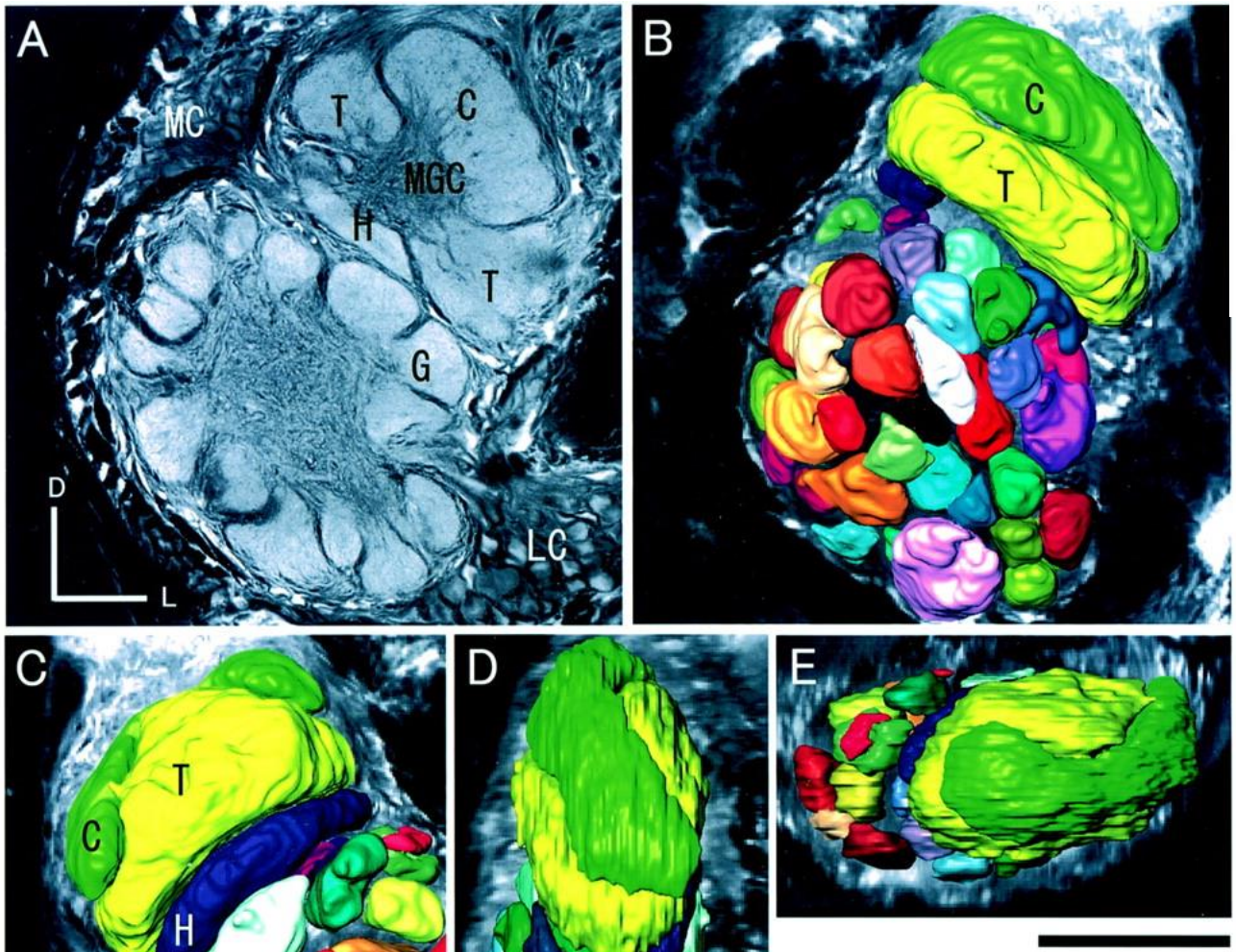
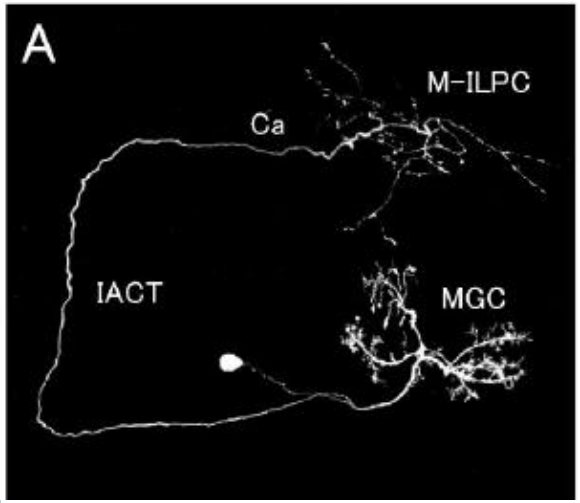
Antennal lobe of male *Bombyx mori*

(*Kanzaki et al. Chemical senses*)





In males an important part of the antennal lobe, the MGC contains the synapses involved in 'Phero' signal processing

Antennal lobe of the male silkworm moth, *Bombyx mori*. (A) A confocal slice image of the LY-stained AL. Three prominent subdivisions of the MGC, the cumulus (C), toroid (T) and horseshoe (H) are shown. (B–E) 3-D reconstruction of the MGC from four different views are shown. Frontal view (B), posterior view (C), side view (D), and dorsal view (E). Scale bar = 100 µm. Direction: D, dorsal; L, lateral.

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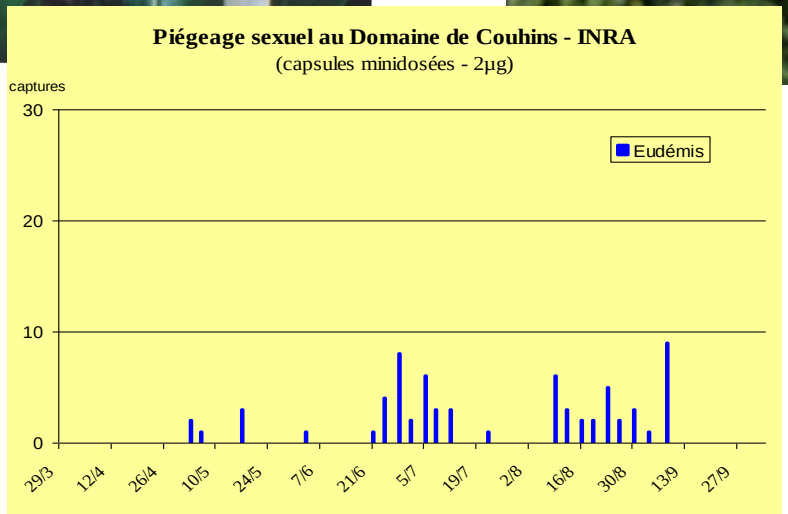
First application of pheromones in crop protection: sexual trap for monitoring



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First publications evoking mating disruption (MD)

Beroza M. ,1960- Insect attractants are taking hold. Agricultural Chemistry, 15: 37-40

Wright R.H., 1964- After pesticides- what ? Nature, 204: 121-125,

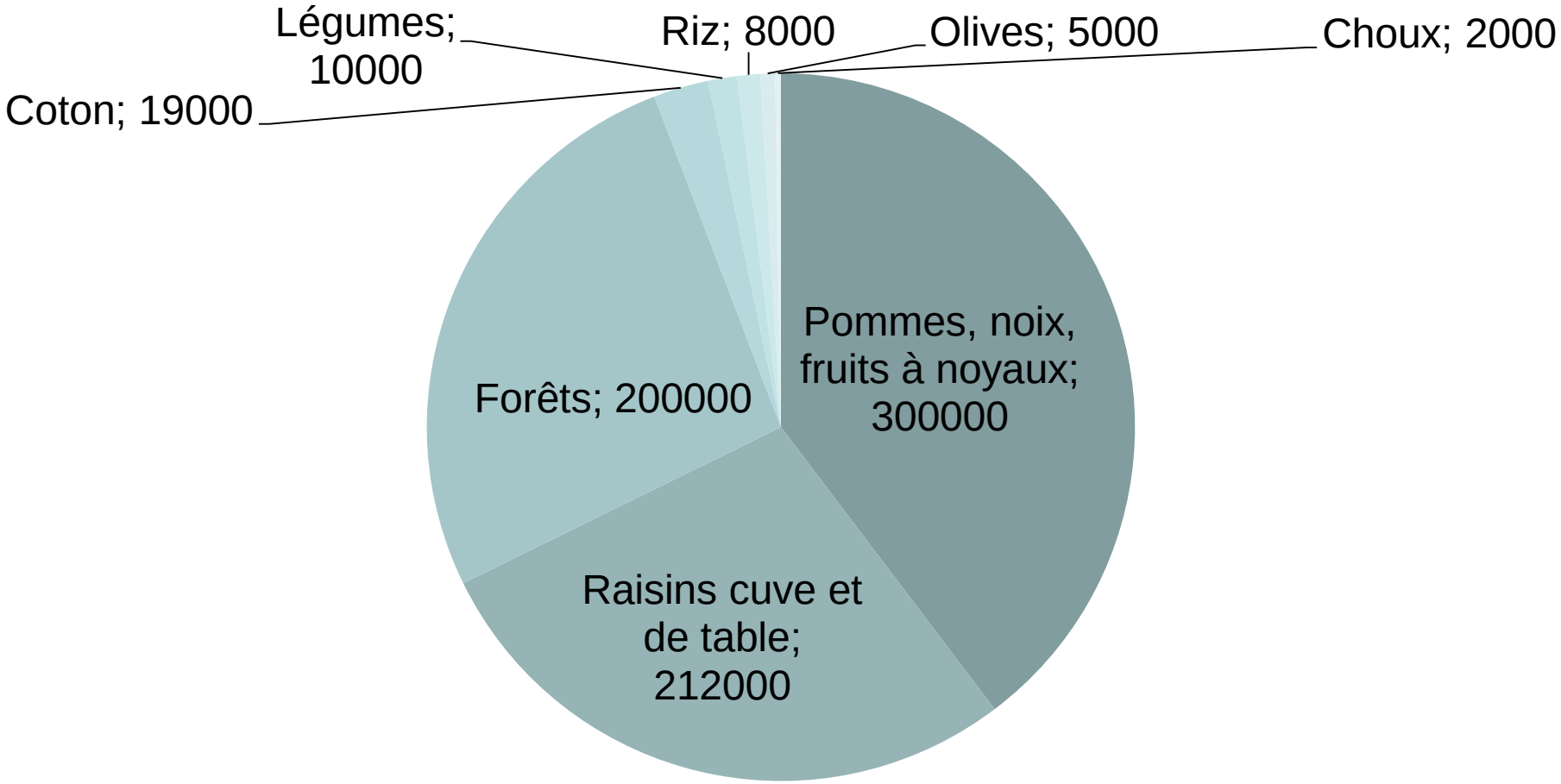
Wright R.H., 1964- Insect control by non toxic means, Science, 144: 487,

First trials of MD against Lepidopteran pests

Acceptable levels of control of the peach moth (*Cydia molesta*) in Australia at a large scale

Rothschild G.H.L.,1975- Bull. Entomol. Research 65: 473-490

Worldwide crop areastreated with : 756.000 ha



Less than 0,05 % of world crop areas, except forests

Data Shin Etzu, 2011

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Vineyards areas under MD in France (total 850.000ha)

in 2010: *ca.* 20.000 ha

2017: > 54.000 ha

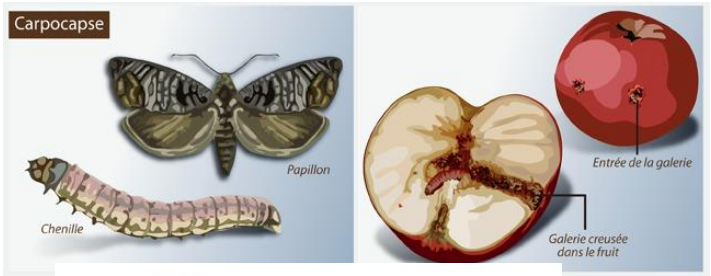
In Europe, currently 3 registered dispensers (passive dispensers and puffer)

Cost (without labour) : from 170 to 240 € x ha^{-1}

Gypsy moth (*Lymantria dispar*), Bombyx disparate

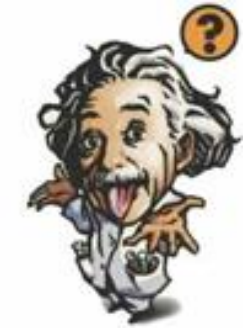
Rice Striped Stem Borer (*Chilo suppressalis)*

Fruit tortricids ex: Codling moth, grape moths oriental fruit moth



Ver rose du cotonier (*Pectinophora gossypiella*)

Mediterranean corn borer / Sésamie du maïs (*Sesamia nonagrioides*)



How works MD in males Lepidoptera?

Principle is based on permeating the air above the crop with enormous doses of sex pheromone, Most of the time, dispenser release the synthetic major constituant at a technical purity. The pheromone is formulated with solvent and often with retardants.

Pheromone can be dispensed using numerous sources (dispensers), e.g. 500 x ha-1 in grapes; or less puffers (see later in the talk), Spraying uniformously pheromone on the foliage is also possible. Leaf cuticule as a wax layer adsorb the pheromone and release it later on.

MD efficiency relies on different effects:

1- Concentration gradients become unconsistent or do no exist anymore.
2- Olfactory fatigue in air saturated by odour (peripheral or central neural adaptation)
3- Dispensers lure the males engaging them in false trails following
4- However, females also detect 'phero' , and may leave saturated environments (recently shown and less studied), See Harrari et al, 2011 Evolution 65: 1572-1582

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An emiting female *Lobesia botrana*

Une femelle >>> quelques nanogrammes à qq centaines ng de phéromone

1 diffuseur (Rak) (250-350mg) pheromone E7Z9-12 Ac equiv. 3-4 x10^7 femelles

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Picture S. Rauscher

To achieve Mating disruption in crops, we need cheap pheromone and dispensers…

… To increase efficiency in future, the quality of the odorant cloud and its 'bevavior in space has to be better studied

Efforts has to be put on:

- Measuring concentration in many places
- Understanding aerodynamics in the crop
- Modelling and controlling concentrations
- Studying the climatic factors (temp. hygrometry)

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Technology of dispensers has progressed a lot in the passed years

≈

Hand-applied reservoir dispensers

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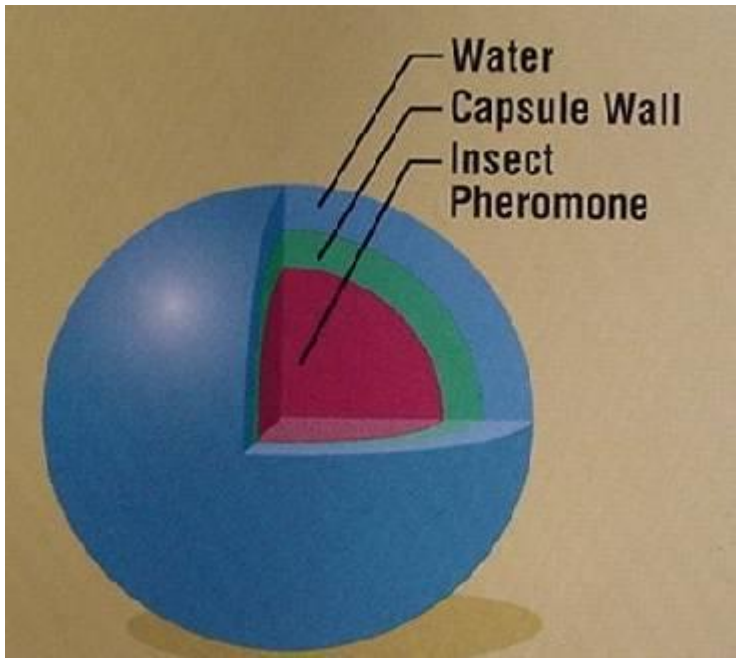
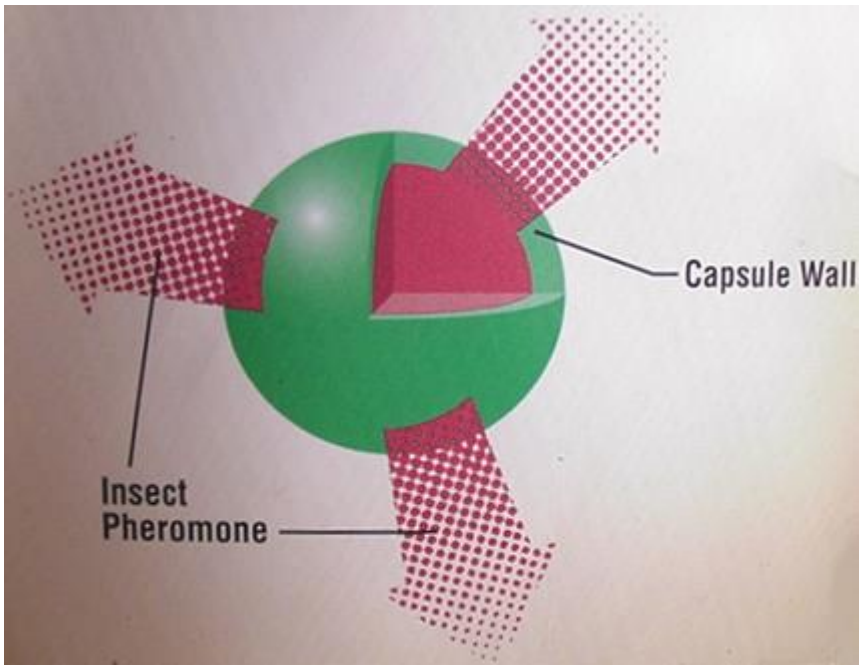
Pictures: L Mattedi, S Rauscher, D Thiéry, unknown internet

Aerosol emitters

Encapsulation in micro caps is also an issue





Advantages :

1- Sprayable on the crop (using classical spray technologies).
2- Multiple sources of release (also use the leaf surface charcteristics)
3- Several applications in the growing seasons allows amount adjustment

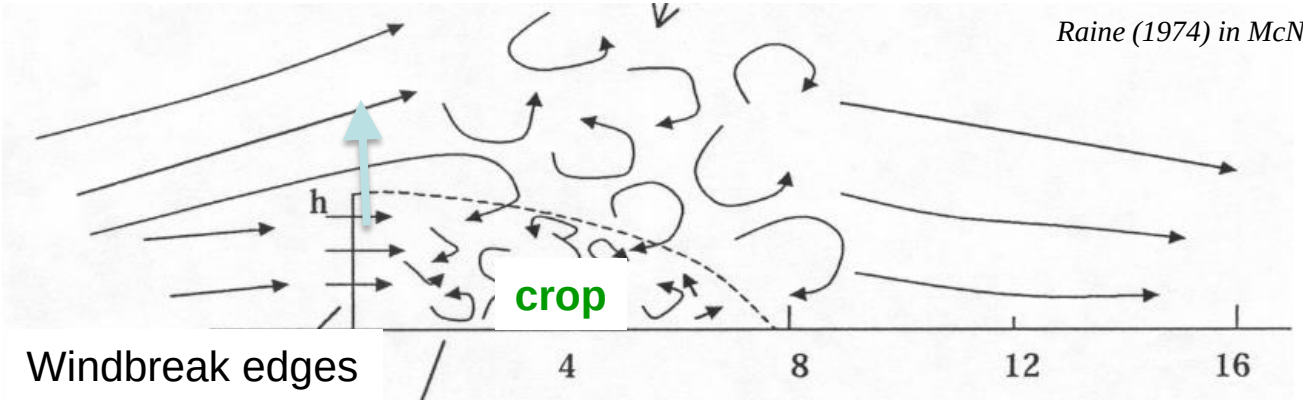
… but only few formulations (3 M company against *Endopiza viteana*, M2I).

MD efficiency relies on the quality of a reliable 'phero cloud' were the pest behaves:

high concentration stable in time and space

e.g. failures may arise in regularly windy vineyards and also sloppy ones:

- **In strong wind, dispensers life duration is reduced (even more critical in hot climates)**



Raine (1974) in McNaughton, (1988)

- **Lepiodptera 'phero' has a tendency to fall on the ground >>> sometimes critical to maintain the optimal concentration where pests behave.**

Case study: grapes,
in European viticulture four main tortricid pests


Eudémis, cochylis, eulia et pyrale



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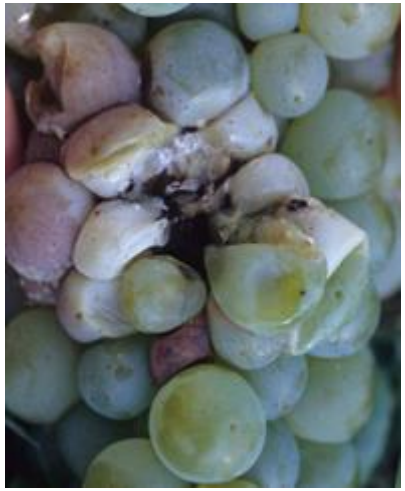
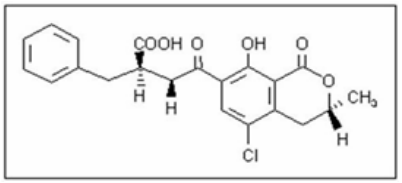
Pictures from Thiéry, 2005

Most damaging are *Lobesia botrana* and *Eupoecilia ambiguella*

Direct damages by larvae which also induce fungi damages



In increasing numbers of mediterranean vineyards *L. botrana* is linked to *Aspergillus (carbonarius, niger)* growth which produces **Ochratoxins**.



Ochratoxin A is a carcinogenic mycotoxin can be found in red wines in cases of heavy attack by black molds. Its amount in wine is regulated in EU and US,

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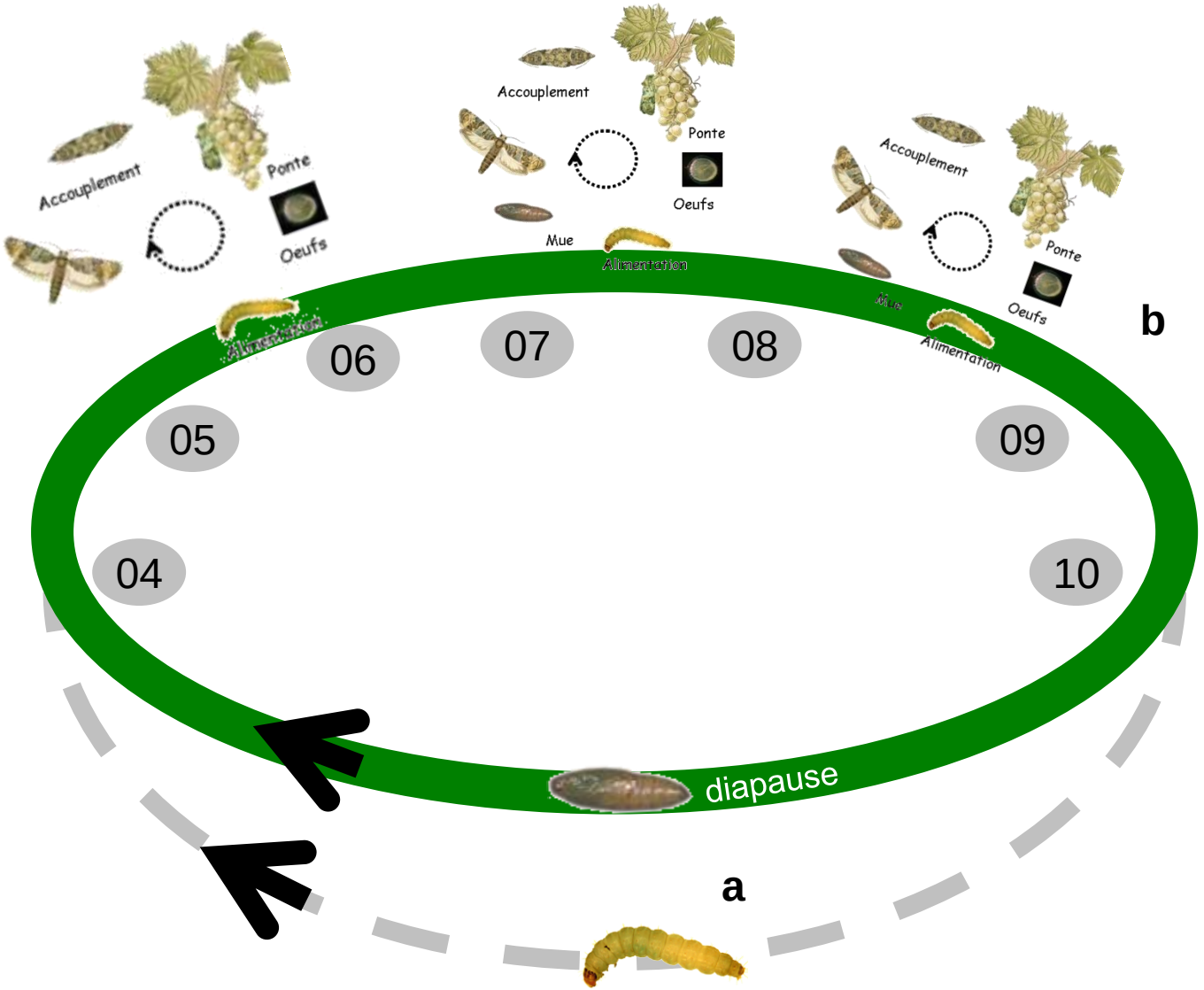
Clichés D Thiéry, D Blancard INRA



Several cycles per year (from 3 to 5 in L. botrana may cause very heavy populations)

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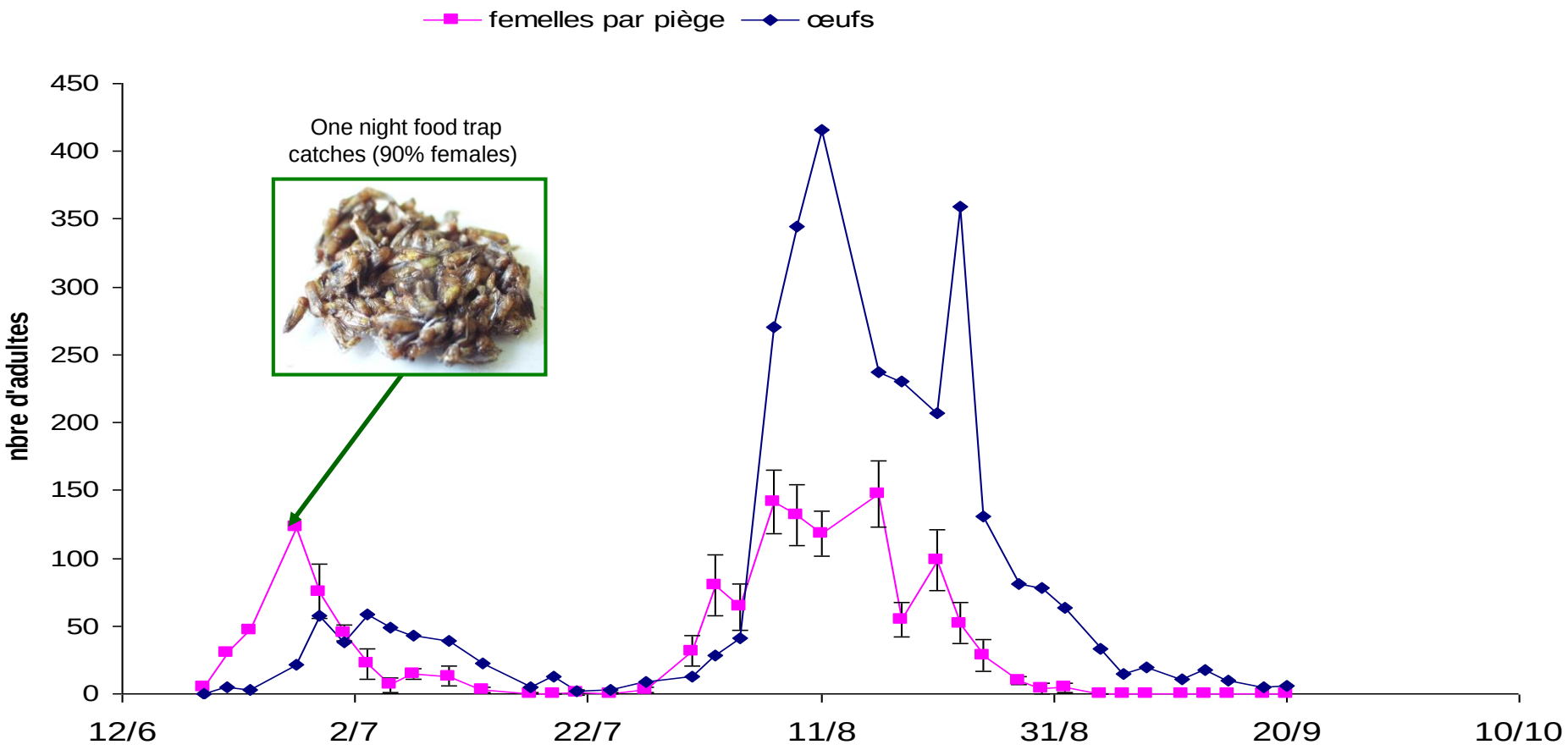
Very strong populations of *L. botrana* can occur

In 2006, this very famous Château of Medoc received 6 insecticide teatments but was damaged 14 larvae per bunch in average at harvest. Only 10hl per ha was vinified (normal yield is 60-70hl)



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Autres temps, une lutte sans relâche

In the Bible (new testament): ‘Dieu vous punira en envoyant des vers rouges entourant de fils vos raisins qui seront détruits’. *(Translation by R Roehrich),*

The so called red worm (*Eupoecilia ambiguella)* was one of the divine scourge with locusts).

1842

HISTOIRE

DES

INSECTES NUISIBLES A LA VIGNE,

ET PARTICULIÈREMENT

DE LA PYRALE

QUI DÉVASTE LES VIGNOBLES DES DÉPARTEMENTS

de la Côte-d'Or, de Saône-et-Loire, du Rhône, de l'Hérault, des Pyrénées-Orientales, de la Haute-Garonne, de la Charente-Inférieure, de la Marne et de Seine-et-Oise;

AVEC L'INDICATION DES MOYENS QU'ON DOIT EMPLOYER POUR LA COMBATTRE.

PAR

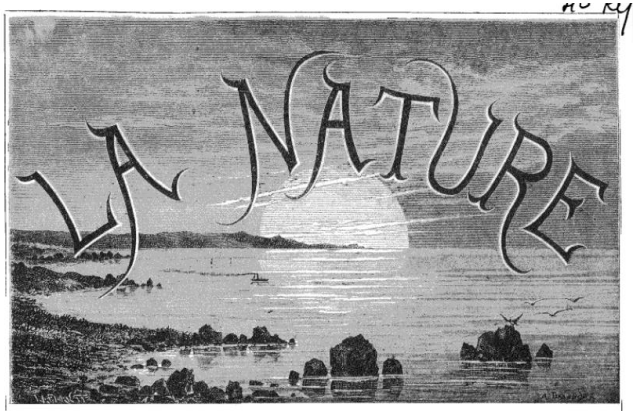
M. VICTOR AUDOUIN,

OUVRAGE PUBLIÉ

SOUS LES AUSPICES DE M. LE MINISTRE DE L'AGRICULTURE ET DU COMMERCE,

ET DES CONSEILS GÉNÉRAUX DES DÉPARTEMENTS RAVAGÉS.

1877



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ET DE LEURS APPLICATIONS AUX ARTS ET A L'INDUSTRIE

JOURNAL HEBDOMADAIRE ILLUSTRÉ

RÉDACTEUR EN CHEF

GASTON TISSANDIER

1912

RAPPORT SUR LES TRAVAUX ACCOMPLIS

PAR LA

MISSION D'ÉTUDE DE LA COCHYLIS

ET DE L'EUDÉMIS

PENDANT L'ANNÉE 1911

PAR

PAUL MARCHAL

Directeur de la Station entomologique de Paris,
Professeur à l'Institut national agronomique.

PUBLIÉ

SOUS LES AUSPICES DU MINISTÈRE DE L'AGRICULTURE

Avec 60 figures dans le texte et deux planches en couleurs

PARIS ET LIÉGE

LIBRAIRIE POLYTECHNIQUE, CH. BÉRANGER, ÉDITEUR

1912

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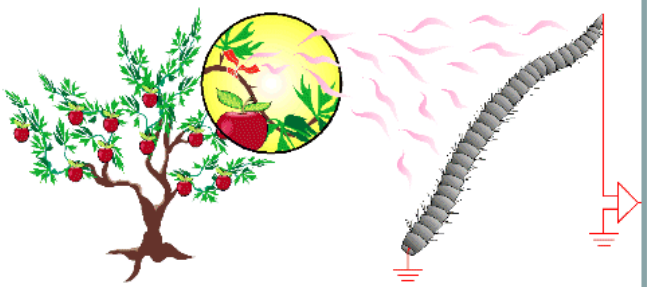
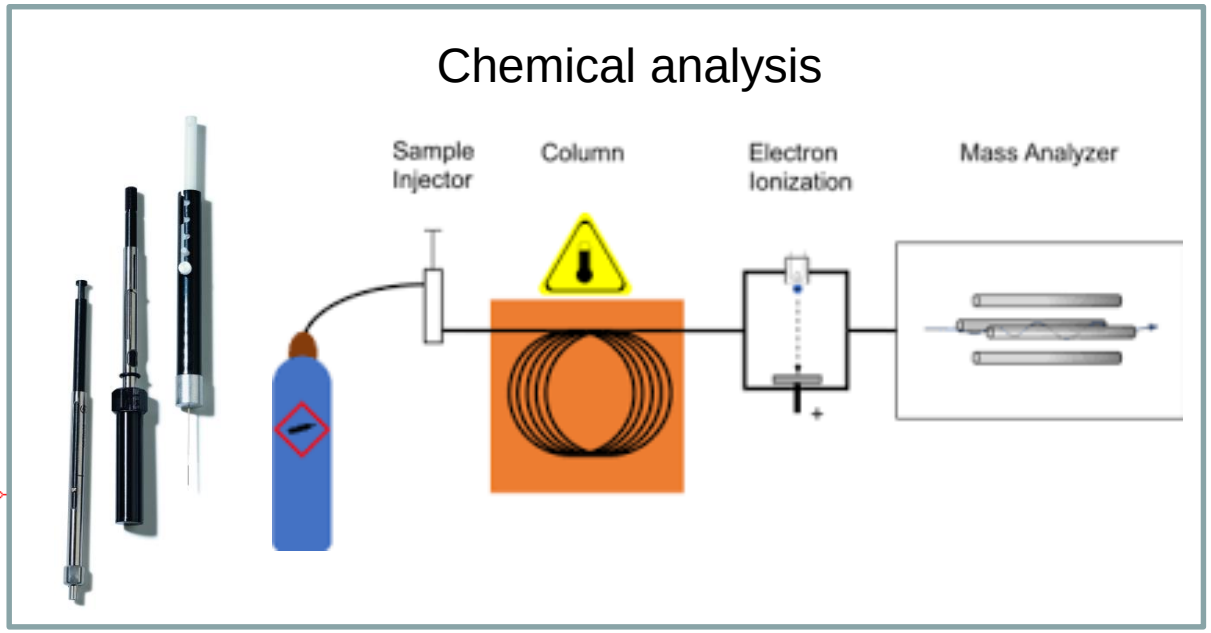
save

santé & agroécologie du vignoble

One century ago, many hard working people were required to control these pests

Currently, measuring phero concentration in a crop is expensive, time consuming, and requires skills.





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Take home messages

Pheromone based methods against pests, and MD are promizing alternatives to classical insecticides.

Analytical chemistry of pheromones made impressive progress.

Targetted behaviors and neurophysiology implicated in mating disruption received important and excellent science.

However the knowledge beyond the diffusion in the air, and how a cloud of pheromone behaves in crop conditions (in space and time) is still challenging.

Aerodynamics and mathematical modelling will undoubtly help in that.

Spatial stable and high concentrationsare needed to avoid so called ‘white areas’ in which adults can easily mate.

This requests frequent fine scale measures of ‘phero’ concentration (space and time).

Research project Sysnum: PHEROtrack

Design a physical sensor to monitor the pheromone release in vineyards

Obj-1: Get numerous concentration data on the crop

Obj-2: Adjust concentration and thus avoiding 'White areas'

Obj-3: Provide spatial data for modeling pheromone diffusion

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Développer un capteur pour mesurer et piloter la diffusion de phéromones

Merci de votre attention ***>>>> Post Doc Petra Ivaskovic***