BSc – MSc projects in the Groot group (Evolutionary chemical ecology)*
December 2018
**If you're interested in one of these subjects, please email a.t.groot@uva.nl to discuss the possibilities.*

The overarching research aim in the Groot lab is to understand when and how sexual selection may drive differentation between populations, and thus initiate speciation. Our research includes behavioral analyses in lab and field experiments to quantify variation in female and male choice and interactions, chemical analyses to assess variation in female and male sex pheromones, genetic analyses to identify the genetic basis of sexually selected traits, and molecular analyses (qPCR, CRISPR-cas9) to functionally characterize the candidate genes.
For more information, see http://www.uva.nl/profiel/g/r/a.t.groot/a.t.groot.html

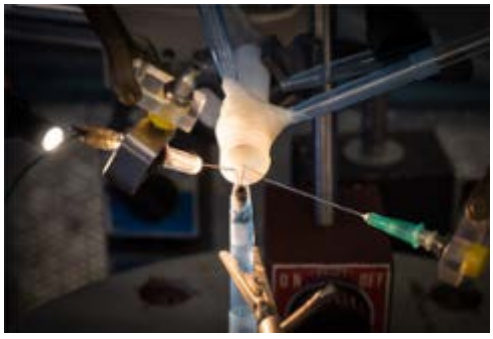
Possible (smaller) research projects for BSc and MSc students are (for full descriptions, see below):
1. Developing an insect-based nose to detect fungal infections in humans (electrophysiological studies and chemical analyses)
2. Ecological immunology: do immunity and sexual attraction trade-off through a desaturase? (experimental manipulation studies, possibly combined with genetic studies)
3. Immunity versus sexual selection: how does sexual attraction in moths trade off with infections of different pathogens?
4. Discover new antimicrobial peptides by analysing the antimicrobial properties of moth pupae (chemical analyses)
5. Circadian rhythm differences between butterflies and moths due to SNPs in clock genes? (genetic analyses)
6. Attraction of *Plutella xylostella* males to sex pheromone traps in the field (field analysis in collaboration with Proeftuin Zwaagdijk)

==========

1. Developing an insect-based nose to detect fungal infections in humans

Type of project:	BSc or MSc internship
Possible length:	3-9 months
Supervisors:	Peter Roessingh (roessingh@uva.nl)
	Teun Boekhout (t.boekhout@westerdijkinstitute.nl)
	Astrid T. Groot (a.t.groot@uva.nl)

Project outline:



Fungal infections in humans are in the majority of the cases caused by at most five pathogenic yeast species. Diagnose tools to determine which of the pathogen species cause the infection are tedious and not straightforward. As these yeast species likely differ in their chemical profiles, it may be possible to distinguish the pathogens based on the volatiles they produce. Insects are known to be much more sensitive in detecting chemical compounds than chemical detection devices, which is the reason that several attempts have been made to develop an 'insect detection device' (Leitch et al. 2013; Strauch et al. 2014). This project aims to

determine whether it will be feasible to develop a 'bionose' to distinguish the main pathogenic yeast species. Preliminary results indicate that this should be possible (Goedhard and Benning, 2018 BSc report UvA).

Techniques:
-Chemical analyses
-GC-EAD
-portable electroantennogram

References:

Leitch O, Anderson A, Kirkbride KP, Lennard C. 2013. Biological organisms as volatile compound detectors: A review. Forensic Science International 232: 92–103.

Strauch M, Luedke A, Muench D. Laudes T, Galizia, CG, Martinelli E, Lavra L, Paolesse R, Ulivieri A, Catini A, Capauano R, Di Natale C. 2014. More than apples and oranges - Detecting cancer with a fruit fly's antenna. Scientific Reports 4: 3576 | DOI: 10.1038/srep03576

==========

2. Ecological immunology: do immunity and sexual attraction trade-off through a desaturase?

Type of project: BSc or MSc internship
Possible length: 3-9 months
Supervisors: Gao Ke (k.gao@uva.nl)
Peter Kuperus (p.kuperus@uva.nl)
Astrid T. Groot (a.t.groot@uva.nl)

Project outline:
The immunocompetence handicap hypothesis (ICHH) states that immunocompetence should trade off with sexual attraction. This basic idea stems from vertebrate research, where testosterone seems to be involved in both immunosuppression and in the brightness of sexual signals. In invertebrates such a direct link has not been found yet. However, our research so far has found that infected moths produce less sex pheromone and the sex pheromone composition shifts towards more saturated compounds instead of the attractive unsaturated pheromone components. These results has led to the following hypothesis that should be tested: Desaturases have a dual role in immune function and in the quality of the sexual signal. In the laboratory, we selected for High vs Low unsaturated sex pheromone compounds, in which the production of unsaturated compounds is almost completely suppressed and now have stable High and Low lines, where the Low line represents a functional knock-out of the desaturase. These lines thus forms the perfect starting point to test this hypothesis. Experiments to conduct will include infection assays in the High and Low lines to determine whether the Low line is more or less immunocompetent than the High line, chemical analyses to determine the effect of infections on the pheromone composition in both lines, and possibly qPCR experiments to assess differential gene expression levels of the desaturases.

Techniques:
-Infection assays
-Chemical analyses
-qPCR experiments

References:

Barthel A, Staudacher H, Schmaltz A, Heckel DG, Groot AT. 2015. Sex-specific consequences of an induced immune response on reproduction in a moth. BMC Evolutionary Biology 15: 282

Groot AT, Schöfl G, Inglis O, Donnerhacke S, Classen A, Schmalz A, Santangelo RG, Emerson J, Gould F, Schal C, Heckel DG. 2014. Within-population variability in a moth sex pheromone blend: genetic basis and behavioural consequences. Proceedings of the Royal Society B 281: 20133054

==========

3. Immunity versus sexual selection: how does sexual attraction in moths trade off with infections of different pathogens?

Type of project: BSc or MSc internship
Possible length: 3-9 months
Supervisors: Gao Ke (k.gao@uva.nl)
Elise Fruitet (e.c.e.fruitet@uva.nl)
Thomas Blankers (t.blankers@uva.nl)
Astrid T. Groot (a.t.groot@uva.nl)

Project outline:

Immune response induction benefits insects in combatting infection by pathogens. However, organisms have a limited amount of resources available and face the dilemma of partitioning resources between immunity and other life-history traits. Since males and females differ in their life histories, sex-specific resource investment strategies to achieve an optimal immune response following an infection can be expected. So far we have found in moths that the female sex pheromone quantity and quality is reduced in infected females, and that the mating success of infected females is negatively affected, which is not the case in males. Recently, we also found indications for a trade-off between attractiveness and immunity; attractive females were more infectious than unattractive females (BSc thesis S. Nichols, D. Muijderman 2018). However, we do not know whether infections affect female and male responses to potential mating partners, although we have preliminary data showing that infected females choose healthy males (MSc thesis Q. Dangh 2018). Since in moths sexual attraction includes female sex pheromones to attract males from a distance and male sex pheromones that are likely used at close range for female choice, we are also interested whether and how infections may affect the male sex pheromone and/or female choice.

Techniques:

-Infection experiments
-Behavioral (mate choice) analyses
-Chemical (pheromone) analyses

References:

Barthel A, Staudacher H, Schmaltz A, Heckel DG, Groot AT. 2015. Sex-specific consequences of an induced immune response on reproduction in a moth. BMC Evolutionary Biology 15: 282

Hosseini SA, van Wijk M, Gao K, Goldansaz SH, Schal C, Groot AT. 2016. Experimental evidence for chemical mate guarding in a moth. Scientific Reports 6: 38567

==========

4. Discover new antimicrobial peptides by analysing the antimicrobial properties of moth pupae

Type of project: BSc or MSc internship
Possible length: 3-7 months
Supervisors: Teun Boekhout (t.boekhout@westerdijkinstitute.nl)
Astrid T. Groot (a.t.groot@uva.nl)

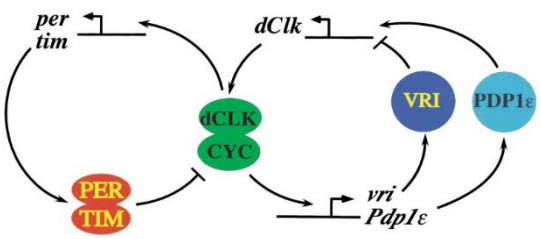
Project outline:
Many night-active butterflies (i.e. moths) feed as caterpillars on various host plants, then pupate in the soil, and after ~ 10 days they emerge as adult butterflies. In rearing moths in the lab on artificial diet, last instar caterpillars dig into the diet to pupate. It is striking how the artificial diet can be very moldy, while the direct environment around the pupal case seems to be completely clean. This raises the question whether pupae excrete antibiotics and/or natural fungicides. This project will determine whether the pupae are indeed as clean as they look by conducting several bioassays (e.g. inhibition zone assays with different media and substrates) with the different life stages of the moths. If these experiments show that pupae are indeed clean, chemical analyses will be done on pupal extracts to characterize the (possible) antibiotics and/or natural fungicides. This project will be conducted in collaboration with the Westerdijk Institute.

Techniques:
-Inhibition zone assays
-Chemical analyses

==========

5. Circadian rhythm differences between butterflies and moths due to SNPs in clock genes?

Type of project: BSc or MSc internship
Possible length: 3-7 months
Supervisors: Sabine Haenniger (shaenniger@ice.mpg.de)
Peter Kuperus (p.kuperus@uva.nl)
Astrid T. Groot (a.t.groot@uva.nl)

Project outline:


The circadian rhythm of behavior has interested many researchers in the past decades, yet amazingly little is known on the evolution of natural variation in circadian rhythms of behavior. Most butterfly species are active during the day, while most moth species show specific activity rhythms in their sexual activities at night[1], some species being sexually active early at night, while others are sexually active late at night[2]. This differentiation has been suggested to have arisen to minimize communication interference between closely related species, as co-occurring and closely related species with overlapping sex pheromone blends show a temporal differentiation in their daily sexual activities. For example, *Spodoptera frugiperda* (Lepidoptera: Noctuidae) consists of two naturally occurring morphologically identical strains that exhibit strain-specific timing of mating in the night[2]: the so-called corn-strain calls, mates and oviposits early at night, while the rice-strain mates late at night. The main candidate gene underlying this

genetic differentiation of this allochronic separation is vrille, which shows butterfly- and moth-specific SNPs (BSc thesis M. Hulswit 2017). The open question is whether this differentiation is vrille-specific or whether other clock genes show a similar pattern. This project is to assess possible genetic variation in identified clock genes in day-active and night-active Lepidoptera in comparison to genes that are unrelated to circadian rhythm.

Requirements: Students need to have some molecular background for this project.

Techniques

-PCR & sequencing genes

-SNP analysis

References

[1]Niepoth N, Gao K, De Roode JC, Groot AT. 2017. Comparing behavior and circadian gene expression between caterpillars, butterflies and moths. *Journal of Biological Rhythms 33: 54-64* doi: 10.1177/0748730417746458

[2]Groot AT. 2014. Circadian rhythms of sexual activities in moths: a review. Frontiers in Ecology and Evolution, section Chemical Ecology 2: 43

[3]Haenniger S, Dumas P, Schöfl G, Gebauer-Jung S, Vogel H, Unbehend M, Heckel DG, Groot AT. 2017. Genetic basis of allochronic differentiation in the fall armyworm. *BMC Evolutionary Biology* 17: 68 (DOI: 10.1186/s12862-017-0911-5)

==========

6. Attraction of *Plutella xylostella* males to sex pheromone traps in the field

Type of project: BSc or MSc internship
Possible length: 3-7 months
Supervisors: Peter Roessingh (roessingh@uva.nl)
Astrid T. Groot (a.t.groot@uva.nl)

Project outline:

The diamondback moth *Plutella xylostella* is the most devastating pest in cabbage worldwide. Now that insecticides need to be banned, it becomes crucial to find alternative and more sustainable methods to combat pest species. Monitoring and mating disruption have been successfully adopted for several moth species (Witzgall et al. 2010). These methods make use of the fact that females attract males by means of a sex pheromone that is emitted from the females from a specialized sex pheromone gland. Every moth species produces its own species-specific sex pheromone (see Pherobase.com) on all identified sex pheromones. However, research over the past decade has shown that there can also be quite some variation within species (e.g. Groot et al. 2009). We also have preliminary results showing that intraspecific variation occurs in the sex pheromone of *P. xylostella*. In this species, host plant adaptation (Löhr B & Gathu 2002) as well as resistance to most insecticides have developed with incredible speed (see e.g. https://en.wikipedia.org/wiki/Diamondback_moth#Pesticide_resistance), which suggests huge genetic diversity and possibly not yet discovered pheromone diversity. This project aims to tests the efficacy of

sex pheromone lures in cabbage fields at the research station Proeftuin Zwaagdijk (see: http://www.proeftuinzwaagdijk.nl/en/). Since the field season starts in May-June, this project will have to be conducted from that period onwards.

Techniques:

-Field experiments

-Preparing sex pheromone lures

-Analysing field traps

-Possibly sex pheromone gland extractions and chemical analyses

References

Witzgall P, Kirsch P, Cork A. Sex pheromones and their impact on pest management. J Chem Ecol 36: 80–100. DOI 10.1007/s10886-009-9737-y

Löhr B & Gathu R. 2002. Evidence of adaptation of diamondback moth, *Plutella xylostella* (L.), to pea, *Pisum sativum* L. Insect Sci. Applic. Vol. **22**: 161–173

Groot AT, Inglis O, Bowdridge S, Santangelo RG, Blanco C, Lopez Jr J, Teran-Vargas A, Gould F, Schal C. 2009. Geographic and temporal variation in moth chemical communication. *Evolution* 63: 1987-2003.