

PULS/CE 28



Photo: Anna Schroll



Max Planck Institute
for Chemical Ecology

Newsletter November 2016



Hawk moths use second nose to evaluate flowers

Floral scent is crucial for successful pollination. *Manduca sexta* hawk moths use their proboscis to smell the floral volatiles when they visit flowers. Olfactory neurons on their proboscises help the moths weigh which flowers to visit ... **p. 3**



Moth takes advantage of defensive compounds

The moth *Heliothis subflexa* benefits from secondary plant components by turning the original defensive function of these compounds into its own advantage. Withanolides, which are present in *Physalis* plants, protect the moth from harmful effects caused by pathogenic bacteria ... **p. 4**



Plant roots in the dark see light

Roots of *Arabidopsis thaliana* plants react directly to light which is transmitted from the shoot to the underground parts. The light activates photoreceptors in the roots and triggers light-dependent growth responses in plants ... **p. 5**





Successful Interdisciplinarity



Alexander Haverkamp is preparing a wind tunnel experiment with moths: The ethologist is interested in how odors in the environment influence the behavior of the tobacco hawk moth *Manduca sexta*. Photo: Anna Schroll

Dear Readers!

In our new issue of PULS/CE, we present two research projects whose exciting results are based on the collaboration between scientists from different disciplines. Our first research highlight describes a successful collaborative effort between two departments of our institute. As I like to explain to visitors, the scientists at the Max Planck Institute for Chemical Ecology study plant-insect interaction from different angles: Plant scientists want to understand phenomena from the perspective of a plant, while insect physiologists are experts at finding out how moths or flies perceive odors and determining how this influences their behavior. This approach urges researchers to “think like a plant!” or “think like an insect!”. When they were studying the flower ecology of the coyote tobacco, researchers from the Department of Molecular Ecology observed that flowers without scent were visited by pollinators as often as scented flowers were. However, the scentless flowers did not produce as many seeds as did the others. Why this is the case could not be answered solely from the perspective of the plant; the scientists also had to study the role of the pollinators.

Collaboration with the Department of Evolutionary Neuroethology resulted in amazing findings. You can read on page 3 about the results.

Another collaborative project with colleagues from South Korea brought together scientists from other disciplines: molecular biologists and optical physicists. In a joint effort, they literally brought light into the darkness and answered a question that had been discussed for decades: are plant roots in the soil able to perceive light? The physicists developed a highly sensitive light detector to measure the intensity of light transmitted in the plant all the way down to the roots. The biologists created plants that were genetically modified so that the light receptors in their roots could no longer be activated. The results of this study, which are summarized in detail on page 5, have just been published as a cover story in the renowned journal *Science Signaling*.

The ability to scrutinize problems from different angles or to deal with them using a variety of strategies may thus be the key to success. We are currently witness an increasing specialization in the scientific world. This makes interactions between the disciplines all the more important; and the most productive results often come from interdisciplinary interactions.

Enjoy learning and reading PULS/CE!

Angela Overmeyer



Felipe Yon studies the flowers of a tobacco plant at the Utah field station. The plant ecologist wants to find out which flowers produce more seeds and why. Photo: Sang-Gyu Kim, MPI-CE





Hawk moths use second nose to evaluate flowers

Flowers without scent produce fewer seeds, although they are visited as often by pollinators as are flowers that do emit a scent. Danny Kessler and Felipe Yon and their colleagues from the Department of Molecular Ecology made this surprising observation when they were studying tobacco plants in the field which had been inhibited in their ability to produce floral odors. But why that was the case was not understood. What role did the pollinators play, especially with regard to their ability to smell flower odors? The ecologists asked their colleagues Markus Knaden and Alexander Haverkamp from the Department of Evolutionary Neuroethology for help. The behavioral scientists were already experienced in performing behavioral assays with hawk moths. For this particular research problem, they developed a so-called y-maze experiment, a proboscis choice test in which a y-shaped tube system is used as an extension of a flower's corolla. The insect must decide which of the two tube endings it will enter with its tongue, otherwise known as its proboscis (see graphic). One of the endings contained floral volatiles, whereas the other did not. This choice test revealed that the moth's proboscis remained much longer in the part of the "flower" that smelled. This result was especially interesting, because the experimental set-up ensured that the antennae were excluded from the odor source. The length of the typical tongue means that the antennae of the moths are quite far away when the tongue is being inserted into the flower. Using molecular biology techniques, the scientists identified the genes which were active on the proboscis. Among these genes were those of two olfactory receptors: the olfactory co-recep-

tor ORCO and the ionotropic co-receptor IR25a. The ORCO gene was active only at the tip of the proboscis, whereas the IR25a gene was also active in the organ's upper parts. These findings show that the proboscis plays a much more important role in olfaction than previously thought. When scrutinizing electron microscopic images of the tip of the proboscis, the researchers discovered a sensillum, a sensory hair, previously unknown in *Manduca sexta*. Further testing revealed that the proboscis sensillum responds to floral volatiles. The perception of floral odor is of crucial importance for pollinators; floral scent is an important indicator of the presence of nectar, because scent provides information about the physiological activity of a flower, and this activity is strongly correlated with nectar production. Moths which correctly locate flower volatiles are also more likely to select flowers which contain enough nectar. On the plant's side, the presence of floral scent increases the fitness of individual flowers for two reasons: first, scenting flowers are more likely to be perceived by pollinators, and second, floral volatiles increase the hawk moths' foraging efforts and thus successful pollination and reproduction.

The evolution of flowers is tightly linked to the evolution of pollinators. This was impressively shown in a further study by Haverkamp et al.: *Manduca sexta* moths also recognize the scent of flowers that best fit their proboscis. The scientists were able to show that the moths acquired the highest energy gain when they visited flowers that matched the length of their proboscis. By learning more about these "postmen of pollen," we learn more about the evolution and functions of plants floral traits. [AO]



Behavioral experiment: A choice assay was set up for *Manduca sexta* moths in order to find out whether floral volatiles influenced how long a moth kept its proboscis inside a flower. At the same time, antennal olfaction was excluded. In the upper part of the y-maze, no odor was introduced; the lower part contained floral volatiles. The moth kept its proboscis much longer in the parts of the flower that had a scent. Therefore the hawk moth seems to be using its proboscis to smell. *Graphic:* Alexander Haverkamp, MPI-CE

Original Publications:

Haverkamp, A., Yon, F., Keesey, I. W., Mißbach, C., Koenig, C., Hansson, B. S., Baldwin, I. T., Knaden, M., Kessler, D. (2016). Hawkmoths evaluate scenting flowers with the tip of their proboscis. *eLife* 5:e15039
Haverkamp, A., Bing, J., Badeke, E., Hansson, B. S., Knaden, M. (2016). Innate olfactory preferences for flowers matching proboscis length ensure optimal energy gain in a hawkmoth. *Nature Communications*. 7:11644





Moth takes advantage of *Physalis* defenses



The larva of the specialist moth *Heliothis subflexa* climbs into the calyx of a *Physalis* seeking shelter.

The calyx, which surrounds the fruit, provides the caterpillar with a safe place to hide from enemies.

Moreover, the fruit contains withanolides, which have antibacterial properties and boost the larva's immune system. Photo: Andrea Barthel MPI-CE

Right below: Andrea Barthel (left) and Hanna Heidel-Fischer (right) with a *Physalis* plant. *Heliothis subflexa* larvae turn defensive withanolides in *Physalis* to their own advantage. Photo: MPI-CE

Original Publication: Barthel, A., Vogel, H., Pauchet, Y., Pauls, G., Kunert, G., Groot, A. T., Boland, W., Heckel, D. G., Heidel-Fischer, H. (2016). Immune modulation enables a specialist insect to benefit from antibacterial withanolides in its host plant. **Nature Communications** 7: 12530

In order to survive and to repel herbivores, many plants defend themselves by producing toxic or deterrent substances. In the course of evolution, many insects have succeeded in adapting to the defensive chemistry of their host plants and thereby circumventing plants' defense mechanisms. However, the plants have also adapted their defensive system to further protect themselves against their enemies, which, in turn, generated counter-adaptations in the insects; biologists refer to this phenomenon as an "evolutionary arms race" between plants and insects. Many insects are plant pests which can be categorized as "specialists" and "generalists." Whereas generalists feed on many different plants, specialists have adapted to one or few closely related plant species as their food.

The moth species *Heliothis subflexa* analyzed in this new study is just such a host specialist. *Physalis* fruits are its favorite food. Researchers from the Department of Entomology compared the effects of withanolides on relative weight gains, survival rates and immune status in two moth species: the specialist *Heliothis subflexa* and the generalist *Heliothis virescens*.

To their surprise they found that only *Heliothis subflexa* benefits from withanolides by increasing larval growth and immune system activity, but its close relative, *Heliothis virescens*, does not. Furthermore, the scientists discovered that withanolides protect the specialist, but not the generalist, from the growth-suppressive effects of an infection caused by the bacterial pathogen *Bacillus thuringiensis*. *Heliothis subflexa* could theoretically profit in two ways

from *Physalis* fruits: First, withanolides display antibacterial and immune stimulant activity; in addition, the *Physalis* fruit is covered by a calyx that creates a so-called enemy-free space.

The researchers aimed to examine the specialization of *Heliothis subflexa* on *Physalis* in the context of ecological immunology, a new field in ecological research. Ecological immunology combines classical studies of the immune system with an ecological perspective to evaluate the costs and benefits of defense against pathogens in the natural environment, and to investigate the manner in which natural selection shapes the immune system.

Further studies will now focus on the mechanisms by which the specialist moth circumvents plant defenses. Moreover, experiments are planned to elucidate the effect withanolides have on the bacterial communities on the plant surface as well as in the gut of *Heliothis subflexa*. [AB/HHF/AO]





Plant roots in the dark see light

Light is not only a source of energy, but also an important signal which regulates many growth processes in a plant in order to adapt it to its environment in the best possible way. Light is first detected by photoreceptors in the shoot of a plant. Physiological processes in the plant are mediated by light-signaling molecules. For more than three decades, scientists have been speculating about whether roots are also able to perceive light. However, until now, this hypothesis has not been proven. Physicists from Korea and biologists from the Department of Molecular Ecology teamed up and combined knowledge and state-of-the-art technologies from both disciplines in order to find out whether plant vascular bundles could act as light optical fibers and transmit light from the shoot to the roots.

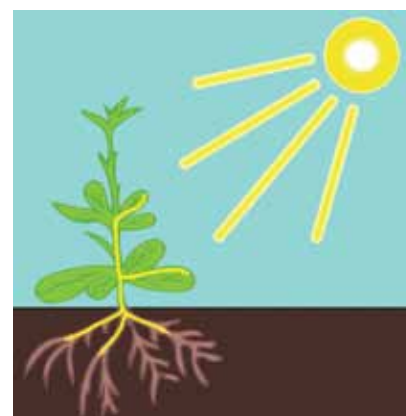
Previous studies had shown that a special photoreceptor in plants, which detects light of the wavelength red/far-red, is, surprisingly, also expressed in the roots. However, it has been unclear how this root photoreceptor was activated. In an interdisciplinary effort, molecular biologists and optical physicists developed a highly sensitive optical detector along with the idea of comparing plants with “blind” roots with those with “sighted” roots. They used plants of the thale cress *Arabidopsis thaliana*, which were genetically modified in a way that the photoreceptor was silenced in the plants’ roots, but not in their shoots. Hence, these plants had “blind” roots.

The scientists grew these modified plants along with control plants; their roots were in the dark soil and their shoots exposed to light, just as in nature.

The optical detector system was used to measure light which was transmitted in the stem down to the roots. With this approach, the researchers were able to show unambiguously that light is transmitted into the roots via vascular bundles. Although the intensity of the transmitted light was low, it was sufficient to activate the photoreceptors, trigger downstream light signaling, and influence growth in the control plants.

These results are crucial for further research projects. The study proves that roots are able to perceive light, even though they are usually found belowground. Photoreception in the roots triggers a signaling chain which influences plant growth, especially root architecture. There are more photoreceptors in the roots. Until now, the responsibilities of these photoreceptors in the roots has remained largely unknown, along with how these interact with light signals which are transmitted from the shoots.

To show the relevance of this study for plants growing in their natural habitat, scientists want to perform experiments with another plant species, the coyote tobacco *Nicotiana attenuata*. This model plant in ecology is adapted to an extremely strong exposure to light. The researchers propose that the newly found sensory modality of roots enhances the ecological performance of plants in nature by timing of resource allocations for growth, reproduction and defense. [KG/AO]



Light transmitted from the shoot to the roots activates photoreceptors in the roots and triggers light-dependent growth responses in plants. Researchers from the Department of Molecular Ecology and Seoul National University, South Korea, were able to show for the first time that roots react directly to the light which is transmitted from the shoot to the underground parts of *Arabidopsis thaliana* plants. Roots can thus effectively adapt plant growth to light conditions in the environment. Graphic: Rakesh Santhanam, Angela Overmeyer, MPI-CE

Original Publication: Lee, H.-J., Ha, J.-H., Kim, S.-G., Choi, H.-K., Kim, Z. H., Han, Y.-J., Kim, J.-I., Oh, Y., Fragoso, V., Shin, K., Hyeon, T., Choi, H.-G., Oh, K.-H., Baldwin, I. T., Park, C.-M. (2016). Stem-piped light activates phytochrome B to trigger light responses in *Arabidopsis thaliana* roots. **Science Signaling**. Vol. 9, Issue 452, pp. ra106





A Vietnamese stick insect, *Ramulus artemis*, climbing a houseplant at the Max Planck Institute for Chemical Ecology. The insect belongs to one of six stick insect species in which the researchers isolated pectinase genes. Photo: Matan Shelomi, MPI-CE

Stick insects produce bacterial enzymes themselves

Many animals depend on their microbiome to digest their food. Symbiotic microorganisms produce enzymes their hosts cannot, and these work alone or together with the animals' own enzymes to break down their food. Many plant-feeding insects need microbial enzymes, such as pectinases, that degrade plant cell walls; yet some insects have overcome this dependency in a surprising way. Researchers from the Department of Entomology found that stick insects make microbial enzymes themselves. From an ancestral gut microbe, the genes for the essential enzymes simply "jumped" as they are to their insect host. The researchers report this newly discovered "horizontal gene transfer" in a paper published in *Scientific Reports*.

Horizontal gene transfer can lead to an astonishing number of new abilities. The human microbiome provides an immense source of potential species-altering proteins. The possibility that genes from microbes living in our guts can suddenly become part of our own genomes and change the course of our evolutionary history is an incredible finding. [MS/AO]

Original Publication: Shelomi, M., Danchin, E. G. J., Heckel, D. G., Wipfler, B., Bradler, S., Zhou, X., Pauchet, Y. (2016). Horizontal gene transfer of pectinases from bacteria preceded the diversification of stick and leaf insects. *Scientific Reports*, 6: 26388



A gopher pulls a branch of *Nicotiana attenuata* into its nest. In nature, the plants are well protected against mammalian herbivory by nicotine. If the plants lack the hormone that regulates nicotine production, they become an attractive food source for many mammals, as shown in experiments with genetically modified plants. Photo: Arne Weinhold, MPI-CE

Jasmonate-deficient tobacco plants attract herbivorous mammals

Coyote tobacco (*Nicotiana attenuata*) produces a potent neurotoxic substance: nicotine. The production of nicotine is regulated by plant hormones called jasmonates. Scientists from the Department of Molecular Ecology, the University of Bern and Washington State University have now demonstrated the importance of jasmonate-dependent nicotine production for the survival of tobacco plants that have been attacked by mammalian herbivores. Through experiments with genetically modified plants that were impaired in their ability to produce jasmonates, the researchers showed that jasmonate-deficiency strongly increases attacks by both insects and vertebrates. Interestingly, insect attack did not significantly affect flower production, and attacked plants were still able to produce seeds, whereas attack by herbivorous mammals had a strong negative

impact on the plants' reproductive ability. The scientists found that nicotine plays a crucial role in this context. Rabbits in particular liked to peel and eat the stems of nicotine-deficient plants. In the outer layers of the stem epidermis, the toxin provides an extremely effective protection.

The researchers want to perform more experiments using mammals as potential herbivores of tobacco plants. They are currently also investigating whether small mammals, such as the wood rat, use nicotine-containing tobacco leaves in their nests as a protection against parasites. [AO]

Original Publication: Machado, R. A. R., McClure, M., Hervé, M. R., Baldwin, I. T., Erb, M. (2016) Benefits of jasmonate-dependent defenses against vertebrate herbivores in nature. *eLife* 5:e13720





The call of the dung

Like many other insects, vinegar flies (*Drosophila melanogaster*) produce pheromones to call their conspecifics to an interesting food source. A research team of the Max Planck Institute for Chemical Ecology in Jena, Germany, demonstrated in a new study that the flies' frass also contains these pheromones. Fruits that have been covered by the insects' fecal excretions seem to be especially attractive to other flies. These fruits probably become a more easily digestible food

after many flies have been feeding on them. The new results are a first step toward understanding the importance of feces in the communication of vinegar flies. [KG/AO]

Original Publication: Keeseey, I., Koerte, S., Retzke, T., Haverkamp, A., Hansson, B. S., & Knaden, M. (2016). Adult frass provides a pheromone signature for *Drosophila* feeding and aggregation. **Journal of Chemical Ecology** 42: 739.



Many vinegar flies have been feasting on this blueberry already. The magnification shows small spots where the flies have left their excrements. The smell of feces makes the fruit even more attractive to the annoying kitchen pests. *Photo: Anna Schroll*

Choosing a mate: It's the brain, not the nose, that knows

How does a male moth find the right sort of female for mating, when two similar types of pheromones are luring it? In many species, differences in the antennae used by the male to smell these perfumes are responsible for its choice. But in the European corn borer (*Ostrinia nubilalis*), changes in the male's brain seem to dictate the choice between two types of available females, as shown by researchers from the University of

Amsterdam, the Swedish University of Agricultural Sciences, and the Department of Entomology [DGH/AO/KG]

Original Publication: Koutroumpa, F. A., Groot, A. T., Dekker, T., Heckel, D. G. (2016). Genetic mapping of male pheromone response in the European Corn Borer identifies candidate genes regulating neurogenesis. **PNAS**, doi: 10.1073/pnas.1610515113



A male and female moth of the European corn borer *Ostrinia nubilalis*. *Photo: Melanie Unbehend, MPI-CE*

Bill S. Hansson receives the International Ellis Island Medal of Honor and an honorary doctorate from the Swedish University of Agricultural Sciences

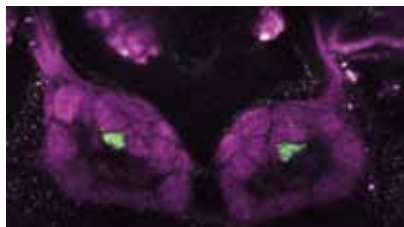
In a ceremony on May 7, 2016, Bill S. Hansson received the International Ellis Island Medal of Honor on Ellis Island, New York, for his contributions to international scientific cooperation and as a global leader in neuroscience research. In addition, the Swedish University of Agricultural Sciences (SLU) awarded him an honorary doctorate on October 8, 2016.

Bill Hansson was Professor of Chemical Ecology at SLU in Alnarp from 2001–2007 before he was recruited to head the Department of Evolutionary Neuroethology by the Max Planck Society. He "has put and will put SLU and the subject of chemical ecology on the scientific world map, now and in the future," the award letter reads. [AO]



Bill S. Hansson wearing the Ellis Island Medal of Honor. *Photo: National Ethnic Coalition of Organizations (NECO)*





Antennal lobe of a vinegar fly: The spherical structures are the olfactory glomeruli. Glomeruli are neuronal networks that are clearly visible in the image as all synapses are marked with a purple-magenta color. The sensory neurons of one particular glomerulus are marked with the photoactivatable green fluorescent protein (PA-GFP).

Fluorescence microscopic image: Veit Grabe, MPI-CE

Olfactory glomeruli have a unique structure

Scientists from the Research Group Olfactory Coding and the Department of Evolutionary Neuroethology have now quantified and mapped the functional units of the olfactory center in the brains of vinegar flies (*Drosophila melanogaster*) responsible for the perception of odors. They found out that the so-called olfactory glomeruli in the antennal lobe, the insect analogue of the olfactory bulb of mammals, differ from each other in their architecture. The morphology and the structure of these spherical brain units provide information about the ecological relevance of the odors they process, especially with regard to the flies' odor-guided behavior. The complete quantitative mapping of all olfactory sensory neurons linked together in

each olfactory glomerulus created an important basis for a better understanding of the role and function these units in the olfactory system have, especially with regard to the processing of behaviorally relevant odors. In all likelihood, these new insights are not limited to the vinegar fly and may also apply to other animals or even humans, and therefore have far-reaching significance. [KG/AO]

Originalveröffentlichung:

Grabe, V., Baschwitz, A., Dweck, H. K. M., Lavista-Llanos, S., Hansson, B., Sachse, S. (2016). Elucidating the neuronal architecture of olfactory glomeruli in the *Drosophila* antennal lobe. *Cell Reports*, 16 (12), 3401-3413



Sandra Irmisch and her poplar trees.
Photo: Tobias Köllner, MPI-CE

Sandra Irmisch receives the Otto Hahn Medal of the MPG

Plants emit odors to defend themselves against attacks by herbivorous animals. Scientists often talk about "the plants' cry for help." Biochemist Sandra Irmisch has studied the poplars' cry for help. For her PhD thesis she investigated the

molecular basis of chemical volatiles poplars emit as a response to infestation by pest insects. The Max Planck Society awarded her the Otto Hahn Medal for an outstanding scientific dissertation. [AO]

Upcoming Event:

20 Years



Max Planck Institute
for Chemical Ecology

20 Years of Cutting-Edge Research in Chemical Ecology

The Max Planck Institute for Chemical Ecology will celebrate its 20th anniversary!

In 1997, the Max Planck Institute for Chemical Ecology opened its doors. Today a leading institution in the field, it is known worldwide for its interdisciplinary research in chemical ecology.

We will celebrate the 20th birthday of our institute on **September 21-22, 2017**.

Details of the program will be announced in due time.

www.ice.mpg.de

