Surviving with symbiotic bacteria
Firebugs need bacterial symbionts to survive on plant seeds as their sole food source. Symbiotic bacteria constitute a key factor not only in the ecological success of firebugs but also in the pest status of cotton strainers ... p. 4

STOP: Avoid rotten food!
Consuming putrid food can be lethal as it allows bacterial pathogens to enter the digestive system. To detect signs of decay is one of the main functions of the sense of smell. The neural mechanisms underlying an escape reflex in fruit flies activated in order to avoid eating rotten food have now been decoded ... p. 3

Not without my microbes
After metamorphosis, European forest cockchafers benefit from the same bacterial symbionts as are found during their larval stage. Microbes in their guts help them to digest their woody food, such as lignocelluloses and xylans ... p. 5
Dear Readers!

After the new Schneiderhaus facility (see PULS/CE 20) started operating, the MPI for Chemical Ecology honored another pioneer in chemical ecology by naming new office space after Thomas Eisner: the Eisnerzimmer. Over the last year, the former library archive was transformed into an open office area where up to 36 junior scientists are able to work. People who enter the room are pleasantly surprised by its bright, friendly atmosphere. You can look out through glass walls and windows on three sides, from the back of the building to a conservation area and even further into the woods. Shannon Olsson, who did her PhD from 2000-2005 under the supervision of Thomas Eisner, is impressed: “There is probably no room in the entire institute which is more appropriate to name after Tom, because this is the room with the most windows. Looking out into nature was always inspiring for him.”

Olsson, who heads a project group in the Department of Evolutionary Neuroethology, often thinks about the most important advice Thomas Eisner gave her – advice she now tells her own students: “If you want to study an insect, a plant or a microbe, you have to think like the insect, the plant or the microbe.” The special legacy Eisner left to the discipline of chemical ecology is his attitude that tracking down the secrets of nature is best done by precisely observing natural phenomena. Despite rapidly developing technologies and methods in molecular biology, natural history and field observations are still crucial.

Melkamu Woldemariam from Ethiopia also shares this opinion. He is about to finish his PhD studies on the identification of novel regulatory mechanisms involved in plant-herbivore interactions in Nicotiana attenuata (see page 6). Much of his work is based on experiments carried out in the plant’s natural habitat.

Enjoy reading PULS/CE and enjoy the wonderful world outdoors!

Angela Overmeyer

The Eisnerzimmer provides room for scientific exchange. Thirty-six young scientists can work here. The room was named after entomologist and ecologist Thomas Eisner (1929-2011), one of the fathers of chemical ecology.

Above: PhD candidate Melkamu Woldemariam at his desk in the Eisnerzimmer.

Right below: Beng Soon Teh from Malaysia and Pol Alonso from Spain, two new PhD students in the Department of Bioorganic Chemistry, talking to Shannon Olsson, who completed her PhD under Eisner’s supervision in 2005.

Photos: Angela Overmeyer/MPI-CE

Eisner’s heirs

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Literature:

STOP: Avoid rotten food!

A shocking number of fatalities occurred in Germany in spring 2011; linked to fenugreek sprouts contaminated with EHEC bacteria, the fatalities showed that it is crucial for every foraging organism to be able to distinguish between fresh food and that infested by pathogenic microbes. Often, visual signs allow us to avoid putrid food. A more direct and common signal, however, is the perception of certain odors released by hazardous microbes. If these odors are detected, the food item is directly discarded, a behavior which can save lives. But which neural functions underlie the capacity to make such judgments? What does the path from the odor molecule and the olfactory receptors into the brain and finally to the reaction of the animal look like?

The genetically well-characterized fruit fly (or vinegar fly) *Drosophila melanogaster* and related species offer perfect systems in which to study such questions. The flies typically feed on yeast growing on rotting fruit and must be able to distinguish between “good” and “bad” microbes. Experiments showed that flies consuming pathogenic bacteria or fungi died immediately. The odor substance geosmin – a smell quite familiar to us and usually associated with the strong scent of wet soils – is known to be produced by several fungi and bacteria and may trigger deterrent behavior.

Scientists in the Department of Evolutionary Neuroethology have now found that *Drosophila* antennae are even more sensitive to geosmin than previously thought. Electrophysiological experiments revealed that a single neuron type, labeled ab4B, responded to geosmin. These neurons carry the specific receptor Or56a, which reacts exclusively to geosmin. Optical imaging of the *Drosophila* brain revealed another interesting result: From the approximately 50 glomeruli that constitute the antennal lobe, the olfactory center of the flies, only one, labeled DA2, was activated by geosmin. DA2 was found in the same region as other glomeruli that are involved in aversive behavior triggered by odors.

The geosmin stimulus, mediated through the dedicated neural line, not only causes the flies to stop or move away from the odor source, it also overrides all other odors offered simultaneously, even highly attractive ones, such as vinegar or fruit scents. Clearly, flies are programmed to avoid geosmin even in a blend of different smells and will not be tempted to consume pathogens accidentally. Geosmin is sufficient and necessary to elicit the avoidance behavior. [BSH/JWK]

Geosmin, the typical smell of toxic bacteria and mold, activates a hardwired avoidance response in fruit flies. A super-sensitive and completely dedicated neural line, from olfactory receptor via sensory neuron and primary brain neurons, is activated as soon as the tiniest amount of geosmin is in the air. This stimulus overrides all other food odor signals, irrespective of how attractive they are on their own. Picture: Marcus Stensmyr, MPI-CE

Original Publication:
Surviving with symbiotic bacteria

Original Publications:

Although the red- and black-colored firebugs that are ubiquitous under linden trees in Central Europe have no impact on humans, their African, Asian, and American relatives, the cotton stainers, are serious agricultural pests. Scientists of the Max Planck Research Group Insect Symbiosis have discovered that these bugs need bacterial symbionts to survive on cotton seeds as their sole food source. The seeds are rich in toxic metabolites and, in addition, poor in essential nutrients. By using high-throughput sequencing technologies and deciphering almost 300,000 copies of bacterial 16S rRNA genes, the scientists discovered that the bugs cultivate a characteristic community of three to six bacterial symbionts in a specific mid-gut region. The symbionts are transferred to the eggs by female bugs, and the hatchlings later take them up by probing the egg surface. Bugs from different localities and even across different species showed very similar microbial communities, indicating that the bugs have been associated with their symbionts over millions of years.

To find out whether the bacterial symbionts help the bugs to survive on the plant seeds as their sole food source, the researchers performed a simple yet elegant experiment: They dipped bug eggs into bleach and ethanol and thereby killed the microbial community on the surface without harming the developing embryo itself. Some of the eggs were then re-infected with a mixture of bacteria from an adult bug’s gut, while others remained symbiont-free.

Interestingly, the symbiont-free individuals showed markedly higher mortality, needed longer to develop into adults, and produced fewer offspring than bugs that had their native symbionts. Thus, symbiont-free bugs showed clear signs of malnutrition, although they were fed on the same plant seeds as their symbiont-bearing counterparts. This strongly indicates an important contribution of the bacteria towards host nutrition. Surprisingly, exchanging bacterial communities between firebugs and cotton stainers also reduced the fitness of both species, indicating that – despite their similarity – the symbioses are highly specific.

Firebugs and cotton stainers are ideal model systems to address fundamental questions in insect symbiosis, because their microbial communities can be manipulated and exchanged, and host fitness can be subsequently measured. Detailed knowledge about how insects interact with microbial symbionts is essential for understanding insect physiology, ecology, and evolution. In the case of agricultural pest insects like the cotton stainers, this knowledge may also provide new biological control strategies. [MK/AO]
Metamorphosis is a fascinating process: A caterpillar or larva, feeding on roots below-ground or leaves above-ground (depending on the species), turns into a butterfly or a beetle after a stage of pupation and quiescence. The cylindrical bodies of larvae are quite unspectacular in comparison to the colorful and delicate bodies of butterflies. On top of that, it is usually the larvae that cause the most damage and threaten agricultural and silvicultural yields by feeding on plants. Among these herbivores is the European forest cockchafer (Melolontha hippocastani), a major pest of trees. During the pupal stage, a fundamental transformation starts, a radical internal conversion that changes every single larval organ. The tissues and organs of the larva are converted into the new organs of the butterfly or beetle. But what happens to the gut microbes that are needed for digesting plant tissues and therefore important for the insect’s survival as soon as the larva is transformed?

Scientists of the Department of Bioorganic Chemistry, the Forest Research Center in Freiburg and the Fritz Lipmann Institute in Jena identified more than 300 individual rRNA sequences that were assigned to the different taxa of known classes of microbes. Nine different classes of bacteria were found in the cockchafer gut, among them proteobacteria, actinobacteria, bacilli, clostridia, negativicutes and sphingobacteria. Some are able to digest lignocelluloses and xylans, typical wood components. Interestingly, many classes of bacteria that were identified in the larval midgut were also found – after metamorphosis – in the gut of the adult cockchafer, even though the larval gut completely empties during the pupal stage. Moreover, the gut microbiome of the larvae overlaps only minimally with the microbiome of soil and root material. In other words, most microbes present in the larvae and beetles do not originate from digested food. This means that the forest cockchafer per se, that is, the larva hatching from the egg, already has a basic set of bacterial symbionts with which this insect species may have co-evolved over thousands of years. This result confirms the assumption that all higher organisms, such as plants, insects and other animals (including humans), are equipped with microbial symbionts. Without these beneficial microbes, we could not live and survive; they must be classified as an integral part of our body. [JWK/AO]

**Not without my microbes**

After the winter, from May until June, European forest cockchafers may occur abundantly and infest the first shoots of oak, maple or beech trees. Once a year, the female Melolontha hippocastani lays up to 30 eggs from which larvae (grubs) hatch; these feed underground on the roots of trees during their three- to five-year larval stage. Below: Cockchafer larva feeding on a carrot in the soil. Above: adult beetle. Photos: Erika Arias Cordero, MPI-CE

Original Publication:
The natural habitat of plants is a symphony of chemical-mediated interactions that govern a plant’s communication with other plants of the same or different species, pollinators or attackers. As active members of this complex web of interactions, plants have to be able to tell friends from foes and tailor their interaction modes accordingly. As major providers of ready-made food, plants are attacked by heterotrophic organisms. Apart from mechanical barriers, such as thorns and trichomes, plants deploy a potent chemical arsenal that could reduce the survival of the attacking herbivores or kill them. However, nothing comes without cost. And due to the high cost of production of these defense metabolites, plants have developed ingenious, multilayered mechanisms for economically regulating the duration and extent of production of these compounds.

Our model plant is *Nicotiana attenuata*, a wild tobacco species. In response to attack, *N. attenuata* plants induce the production of toxic secondary defense compounds including nicotine, trypsin protease inhibitors, diterpene glycosides and phenolamides. The biosynthesis of these chemical defenses depends on the activation of the jasmonate signaling cascade; this cascade involves the production of huge pools of jasmonic acid (JA) and its bioactive form, jasmonoyl-L-isoleucine (JA-Ile). As being constitutively defensive is costly to the plants, we asked how *N. attenuata* plants regulate the jasmonate signaling cascade in general and, specifically, the production of herbivore-induced JA-Ile.

We identified a novel enzyme that attenuates the jasmonate burst and, consequently, regulates the production of defense metabolites. When we knocked down the expression of this enzyme, a hydrolase named jasmonoyl-L-isoleucine hydrolase 1 (JIH1), in *N. attenuata* and transplanted the transformed plants to their natural habitat, we observed that the transformed plants accumulated more JA-Ile and were better protected against herbivory than were untransformed plants. Moreover, the transformed plants attracted more egg predators (*Geocoris spp.*) to experimentally attached eggs of the tobacco hornworm *Manduca sexta*. This correlated with the higher production of the volatile organic compounds that attract the predators. These observations suggested that by regulating the jasmonate burst, JIH1 affects plants’ physiology and biochemistry, and, ultimately, the ecological interaction of *N. attenuata* plants with its natural herbivore communities.

Melkamu Gezahagne Woldemariam
Transistor in the fly antenna: Insect odorant receptors regulate their own sensitivity

Highly developed antennae containing different types of olfactory receptors allow insects to use minute amounts of odors to orient themselves towards resources such as food, oviposition sites or mates. Scientists have now used mutant flies and for the first time provided experimental proof that the extremely sensitive olfactory system of fruit flies is based on the self-regulation of odorant receptors. Even very few odor molecules below the response threshold are sufficient to amplify the sensitivity of the receptors, and the additional binding of molecules shortly afterwards finally triggers the opening of an ion channel that controls the fly’s reaction and flight behavior. This means that a below-threshold odor stimulation increases the sensitivity of the receptor and suggests that if a second odor pulse arrives within a certain time span, a neural response will be elicited. **Original Publication:** Getahun, M. N., Olsson, S., Lavista Llanos, S., Hansson, B., Wicher, D. (2013). Insect odorant response sensitivity is tuned by metabotropically autoregulated olfactory receptors. *PloS One*, 8(3): e58889.

Metal ions regulate terpenoid metabolism in insects

Scientists have discovered that an enzyme that catalyzes chain elongation in the terpenoid metabolic pathway is regulated in an unusual way. In the horseradish leaf beetle, *Phaedon cockleariae*, a single enzyme, a prenyl synthase, can trigger the production of two completely different substances depending on whether it is regulated by cobalt, manganese or magnesium ions: iridoids, which are defensive substances the larva uses to repel predators, or juvenile hormones, which control the insect’s development. Hence, insects may have developed an efficient option to channel metabolites into the different directions of terpenoid metabolism by simply using metal ions for control. **Original Publication:** Frick, S., Nagel, R., Schmidt, A., Bodemann, R., Rahfeld, P., Pauls, G., Brandt, W., Gershenzon, J., Boland, W., Burse, A. (2013). Metal ions control product specificity of isoprenyl diphosphate synthases in the insect terpenoid pathway. *Proceedings of the National Academy of Sciences of the United States of America*, 110(11), 4194-4199.

Herbivore defense in ferns

Researchers in the Department of Bioorganic Chemistry have now found that bracken ferns (*Pteridium aquilinum*) do not release any volatiles when they are attacked by herbivores, unlike many of the now dominant and evolutionarily younger flowering plants. Such an emission of volatile compounds may attract the pest insects’ enemies, for example, ichneumon wasps or predatory bugs, which parasitize herbivores. However, volatile emission can be elicited in fern fronds that had been treated with the plant hormone jasmonic acid. This suggests that ferns can in principle mobilize an indirect kind of defense reaction like flowering plants. However, they don’t seem to use this form of defense to fend off herbivores. **Original Publication:** Radhika, V., Kost, C., Bonaventure, G., David, A., Boland, W. (2012). Volatile emission in bracken fern is induced by jasmonates but not by *Spodoptera litoralis* or *Strongylogaster multifasciata* herbivory. *PLoS One*, 7(11): e48050.
Jonathan Gershenzon honored as AAAS Fellow

The American Association for the Advancement of Science (AAAS) has elected Jonathan Gershenzon as an AAAS Fellow in the section of Biological Sciences. With this honor, the AAAS acknowledges Gershenzon’s distinguished contributions to our knowledge of the composition, ecological role and evolution of plant defense compounds. The AAAS is the world’s largest scientific society. It was founded in Pennsylvania in 1848. Since 1880, the AAAS has been publishing the journal Science.

www.aaas.org

Upcoming Events

Around 40 junior scientists will meet on September 5 and 6, 2013, during the 7th Kurt Mothes PhD Workshop on Secondary Metabolism. The event will be held in the lecture hall of the Abbe Center on the Beutenberg Campus. Topics at the meeting will include the latest research developments on bioactive agents from plants and microorganisms. The workshop, which is dedicated to the memory of the famous plant physiologist and natural product researcher Kurt Mothes (1900-1983), has been organized since 1996 jointly by the Leibniz Institute of Plant Biochemistry in Halle and the Max Planck Institute for Chemical Ecology.

www.ice.mpg.de/ext/ceepc2013.html

The 7th Central and Eastern European Proteomics Conference (CEEPC) on Proteomics Driven Discovery and Applications will be held October 13-16, 2013, at the Abbe Center on the Beutenberg Campus in Jena. Aleš Svatoš, who heads the Mass Spectrometry and Proteomics Research Group at the MPI, will be organizing this event for the second time in Jena. Planned symposia include clinical proteomics, high throughput technologies, plant, insect and bacterial proteomics, as well as the imaging of biomarkers.

The 5th Long Night of the Sciences will begin in Jena on Friday, November 29, 2013, at 6:00 p.m. The MPI-CE will open its doors to the public until midnight. The first program highlight has already been confirmed: Prof. Dr. Ernst Peter Fischer (picture on the left), historian of science, will give a public talk about “Romantic Science – 100 years of Bohr’s model of the atom.”

www.sternstunden-jena.de