

articulatory-defined phonetic features, such as lip-rounding or tongue position [11]. The Leonard *et al.* [2] results for signing are surprisingly parallel, despite the dramatic difference in linguistic articulators. Speaking and signing both involve rapid, fine-grain coordination of multiple articulators that together encode abstract linguistic representations of form ('phonemes'). This universal level of structure appears to be supported by the same neural principles and architecture, regardless of language modality. Future research in both linguistics (clarifying the nature of phonological features in sign language) and neuroscience (more controlled mapping of the neural encoding of these features in time and space) will reveal the extent to which phonological structure in human language is modality-independent versus specific to the manual or vocal articulators.

#### REFERENCES

- Brentari, D. (2019). *Sign Language Phonology* (Cambridge University Press).
- Leonard, M.K., Lucas, B., Blau, S., Corina, D.P., and Chang, E.F. (2020). Cortical encoding of manual articulatory and linguistic features in American Sign Language. *Curr. Biol.* *30*, 4342–4351.
- Emmorey, K., McCullough, S., Mehta, S., Ponto, L.L.B., and Grabowski, T.J. (2011). Sign language and pantomime production differentially engage frontal and parietal cortices. *Lang. Cogn. Process* *26*, 878–901.
- Thompson, R., Emmorey, K., and Gollan, T.H. (2005). "Tip of the fingers" experiences by deaf signers: insights into the organization of a sign-based lexicon. *Psych. Sci.* *16*, 856–860.
- Klima, E., and Bellugi, U. (1979). *The Signs of Language* (Harvard University Press).
- Levelt, W.J., Roelofs, A., and Meyer, A.S. (1999). A theory of lexical access in speech production. *Behav. Brain Sci.* *22*, 1–38.
- Emmorey, K., Winsler, K., Midgley, K.J., Grainger, J., and Holcomb, P.J. (2020). Neurophysiological correlates of frequency, concreteness, and iconicity in American Sign Language. *Neurobiol. Lang.* *1*, 249–267.
- Emmorey, K., Mehta, S., McCullough, S., and Grabowski, T.G. (2016). The neural circuits recruited for the production of signs and fingerspelled words. *Brain Lang.* *160*, 30–41.
- San José-Robertson, L., Corina, D.P., Ackerman, D., Guillemin, A., and Braun, A.R. (2004). Neural systems for sign language production: mechanisms supporting lexical selection, phonological encoding, and articulation. *Hum. Brain Mapp.* *23*, 156–167.
- Emmorey, K., Bosworth, R., and Kraljic, T. (2009). Visual feedback and self-monitoring of sign language. *J. Mem. Lang.* *61*, 398–411.
- Bouchard, K.E., Mesgarani, N., Johnson, K., and Chang, E.F. (2013). Functional organization of human sensorimotor cortex for speech articulation. *Nature* *495*, 327–332.

## Insect Host Choice: Don't Put All the Eggs in One Basket

Mohammed A. Khallaf and Markus Knaden\*

Department of Evolutionary Neuroethology, Max Planck Institute for Chemical Ecology, Hans-Knöll-Straße 8, D-07745 Jena, Germany

\*Correspondence: [mknaden@ice.mpg.de](mailto:mknaden@ice.mpg.de)

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Unlike mammals, most insects have no chance to personally take care of their offspring. Insect mothers, therefore, carefully weigh egg-laying options to select an optimal site, which guarantees better survival and fitness for their progeny. A new study in oriental fruit flies reveals that gravid females rely on a bacteria-derived odour —  $\beta$ -caryophyllene — to avoid competition for their offspring.

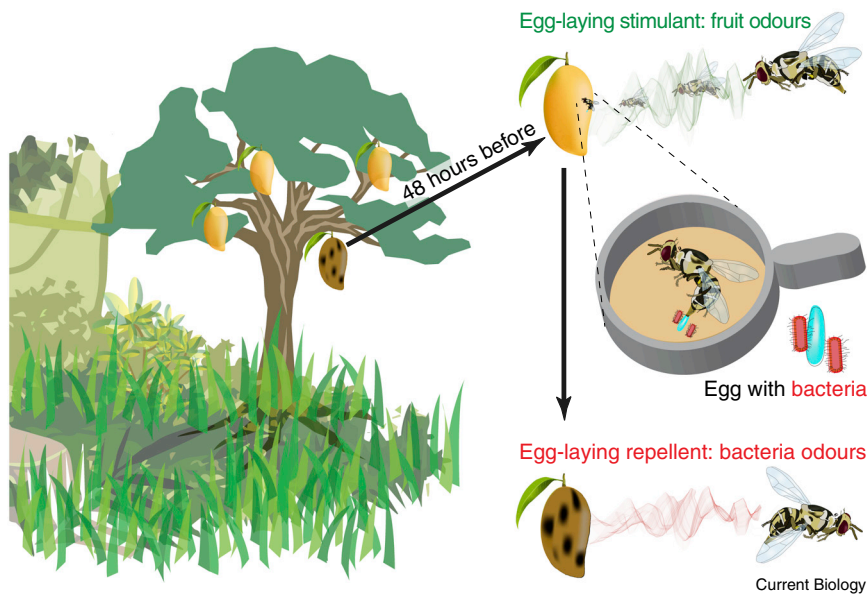
Animals have evolved sophisticated strategies to increase survival and fitness for their progeny [1]. A wide range of different behaviours can be observed throughout the animal kingdom: for example, lactation in mammals [1], active egg guarding against predators in birds [2], egg fanning to increase oxygen access in fishes [3], and active removal of microbes and fungi from eggs in millipedes [4]. It can even reach extreme levels like in some amphibians [5], where the skin of the mother becomes consumed by her offspring (a behaviour known as matrophagy). However, most invertebrates provide no direct care for

their offspring, except a limited amount of yolk in the egg that serves as initial food source and a well-selected oviposition site. How do gravid females evaluate opportunities and threats when choosing their egg-laying substrate? How do they determine the sufficiency of food resources and the presence or absence of potential intraspecific competitors? In this issue of *Current Biology*, Huijing Li, Lu Ren, and colleagues [6] discover a strategy used by the polyphagous oriental fruit fly *Bactrocera dorsalis* to mark its host, unravelling an extraordinary interaction between bacteria, insects, and plant hosts (Figure 1). Their study offers

novel insights into how insect oviposition-related decisions are shaped by tritrophic interactions and extend our understanding of the ecological relationships between bacteria and insects.

Although a few insects are viviparous (the mothers bear their neonates), most are oviparous, that is, they lay eggs, which are unable to move and hence are vulnerable to biotic and abiotic threats. Therefore, selecting an appropriate egg-laying substrate is a challenging task for the gravid insect female. This is especially true for holometabolous insects (those that exhibit a complete metamorphosis),





**Figure 1. The oriental fruit fly avoids laying eggs on fruits that are already infested by conspecifics.**

A female fly chooses its host fruit based on fruit odours. Egg-derived bacteria later govern the fruit-based production of  $\beta$ -caryophyllene. This odour repels further gravid females and hence reduces interspecific competition of the flies.

whose larvae are still relatively immobile and whose chances for survival depend on their mother's decision of where to lay her eggs; for example, an unoccupied oviposition site avoids a shortage of food due to intraspecific competition. One way for the mother to avoid competition for her offspring is to mark her oviposition site with a specific odour — called a host-marking pheromone — which tells other mothers that the place is already taken. This phenomenon was first observed by William Kirby and William Spence ~160 years ago [7], when they noticed that females of the cabbage butterfly preferred to lay their eggs on plants free of eggs. However, it took another 110 years before the first host-marking pheromone was isolated from the Azuki bean weevil [8]. Currently, host-marking pheromones of more than 200 species of parasitoid wasps and 30 species of herbivorous insects have been discovered [9], and many are used as very effective tools in integrated pest-management programs [10].

One of the world's most damaging pests to mark its egg-laying hosts is *B. dorsalis*. Gravid females of *B. dorsalis* infest more than 400 plant hosts [11] and can travel more than 20 miles [12] in search of a suitable fruit in which they lay

eggs between the peel and flesh [13]. Larvae hatch within 1–3 days and then penetrate the flesh, causing the whole fruit to rot [13]. Despite its economic importance, no specific chemical has yet been identified as a host-marking pheromone for *B. dorsalis*. In their paper, Li and Ren *et al.* [6] set out to investigate how gravid *B. dorsalis* females avoid laying eggs in sites occupied by conspecific eggs or larvae. The authors first established a two-choice oviposition preference assay and showed that flies generally avoid oviposition in fruit harbouring conspecific eggs. Surprisingly, the oviposition avoidance is dependent on the age of the eggs, with older eggs being more strongly avoided.

The 'mother-knows-best' hypothesis predicts that insect females should lay eggs at sites that provide the best survival chances to their progeny. To choose these sites, females assess a wide range of signals about the surrounding environment, through the integration of different sensory modalities. Of the sensory cues involved, odours represent reliable long-range signals with which to evaluate substrates. For example, through pheromone-sensing olfactory channels,

gravid females of *Drosophila melanogaster* attract each other to lay eggs on the same site to increase offspring survival through larval cooperation [14]. Likewise, tobacco hawkmoths avoid oviposition in sites occupied with conspecific larvae through sensing acid-derived faeces by olfaction [15]. Through a combination of chemical and electrophysiological experiments, using solid phase microextraction and electroantennography detection, Li and Ren *et al.* [6] analysed chemical profiles of fruit before and after infestation by *B. dorsalis* and tested whether the flies' antennae — the main olfactory organs in insects — would respond to them. Using these techniques, the authors identified  $\beta$ -caryophyllene as the most promising deterrent candidate, as it increased after infestation in all tested fruits (guava, orange, and mango) and could be detected by the flies. Behavioural assays further supported the role of this odour. In a four-arm olfactometer assay, the high concentration of

$\beta$ -caryophyllene found in infested fruit repelled *B. dorsalis* females. Similarly, in a two-choice oviposition assay, gravid females significantly prefer fresh fruit over similar ones perfumed with  $\beta$ -caryophyllene.

Egg deposition is a warning signal of upcoming larval feeding damage. Therefore, plants respond to it through direct or indirect defensive mechanisms that might include herbivory-induced plant volatiles [16]. Gravid herbivorous females can adjust their egg deposition according to these volatiles to avoid intraspecific competition [16]. But, is  $\beta$ -caryophyllene really a plant-defence-related compound that is induced by the laid eggs? In a series of further experiments, the authors showed that not the eggs themselves, but rather the bacteria on the egg surface are the major players behind the increase of  $\beta$ -caryophyllene after infestation. First, using a bacterial fluorescent marker, the presence of bacteria on the surfaces of eggs was visualized. Second, chemical analyses revealed that the  $\beta$ -caryophyllene concentration in fruit increases not only in the presence of eggs, but also when the fruit are treated with bacteria-containing eluent obtained from washing of the eggs. Lastly,

behavioural experiments confirmed that this eluent is sufficient to induce oviposition deterrence in *B. dorsalis*. Egg-laying decisions in several dipteran species are influenced by the presence of bacteria, which produce a blend of volatiles belonging to different chemical classes [17]. One of these volatiles is  $\beta$ -caryophyllene, whose concentration has been reported to rise after the infection of plants with pathogenic bacteria [18]. Therefore, Li and Ren *et al.* [6] hypothesized that increased levels of  $\beta$ -caryophyllene are not attributed to the host plant itself but to the bacteria present on the surface of the eggs.

Bacteria, which are abundant in insects and their ecological niches, play a vital role in mediating the interactions between host plants and insect herbivores. In several tritrophic interactions, bacteria associated with insects or host plants could provide many benefits to eggs and larvae, such as providing nutritional substances and/or antimicrobial compounds [17]. Likewise, insect-associated bacteria could improve insect fitness by detoxifying plant-defensive chemicals [19]. But which are the bacterial players in the oviposition choice of the oriental fruit fly? Through sequencing PCR-amplified ribosomal 16S rRNA, the authors identified, among others, the bacterial genera *Providencia*, *Klebsiella*, and *Enterobacter* on the egg surfaces. Notably, the abundance of *Klebsiella* and *Enterobacter*, but not *Providencia*, increased 48 hours after infestation, correlating with the increased oviposition avoidance of gravid females at that time. Li and Ren *et al.* [6] next demonstrated that *Klebsiella* and *Providencia* spp. isolated from *B. dorsalis* eggs were sufficient to deter oviposition preference in gravid females.

Deciphering the identity and behavioural significance of microbe-induced volatiles is providing new opportunities to control insect pests and host plants. The discovery of the host-marking odour of the oriental fruit fly is intriguing and inspires more pest-control-related questions. Does spraying the hosts with  $\beta$ -caryophyllene reduce *B. dorsalis* infestation? And if so, does the fly develop resistance or habituation towards it? Future field experiments testing the efficiency of this newly

identified chemical will be needed to generalize its applicability to the biocontrol programs of this pest. Moreover, additional research is needed to know the source of these bacteria and whether females transfer them to eggs. It is also important to investigate the plant responses to increased concentration of  $\beta$ -caryophyllene.

The insect nervous system is fascinating with respect to how it governs behaviour, such as egg laying. Oviposition behaviours are sexually dimorphic, and most of the neurons composing the underlying circuits can be expected to be under the control of female-specific genetic loci. Based on this, it should be possible to identify and characterize the olfactory channels, including the odorant receptors, that mediate this oviposition deterrence. It would also be interesting to test whether the role of  $\beta$ -caryophyllene (and the corresponding receptors) in oviposition deterrence is conserved among different *Bactrocera* species. This genus includes more than 70 species that are pests on a wide range of fruit and vegetables [20]. If the novel findings of this new study apply to some or all *Bactrocera* species, the genus might become at least somewhat less scary to the agricultural industry.

## REFERENCES

- Royle, N., Smiseth, P., and Kolliker, M. (2012). *The Evolution of Parental Care* (Oxford: Oxford University Press).
- Moller, A.P., and Cuervo, J.J. (2000). The evolution of paternity and paternal care in birds. *Behav. Ecol.* **11**, 472–485.
- Green, B.S., and McCormick, M.I. (2005). O<sub>2</sub> replenishment to fish nests: males adjust brood care to ambient conditions and brood development. *Behav. Ecol.* **16**, 389–397.
- Kudo, S.I., Akagi, Y., Hiraoka, S., Tanabe, T., and Morimoto, G. (2011). Exclusive male egg care and determinants of brooding success in a millipede. *Ethology* **117**, 19–27.
- Kupfer, A., Muller, H., Antoniazzi, M.M., Jared, C., Greven, H., Nussbaum, R.A., and Wilkinson, M. (2006). Parental investment by skin feeding in a caecilian amphibian. *Nature* **440**, 926–929.
- Li, H., Ren, L., Xie, M., Gao, Y., He, M., Hassan, B., Lu, Y., and Cheng, D. (2020). Egg-surface bacteria are indirectly associated with oviposition aversion in *Bactrocera dorsalis*. *Curr. Biol.* **30**, 4432–4440.
- Kirby, W., and Spence, W. (1863). *An Introduction to Entomology, or Elements of the Natural History of Insects*, 7<sup>th</sup> Edition (London: Longman, Green, Longman, Roberts and Green).
- Oshima, K., Honda, H., and Yamamoto, I. (1973). Isolation of an oviposition marker from azuki bean weevil, *Callosobruchus chinensis* (L.). *Agr. Biol. Chem. Tokyo* **37**, 2679–2680.
- Liu, S., Zhao, B., and Bonjour, E. (2011). *Host Marking and Host Discrimination in Phytophagous Insects* (Dordrecht: Springer).
- Silva, M.A., Bezerra-Silva, G.C.D., and Mastrangelo, T. (2012). The host marking pheromone application on the management of fruit flies – a review. *Braz. Arch. Biol. Techn.* **55**, 835–842.
- Liquido, N.J., McQuate, G.T., Birnbaum, A.L., Hanlin, M.A., Nakamichi, K.A., Inskeep, J.R., Ching, A.J., Marnell, S.A., and Kurashima, K.S. (2017). A review of recorded host plants of oriental fruit fly, *Bactrocera (Bactrocera) dorsalis* (Hendel) (Diptera: Tephritidae), version 3.0, USDA Compendium of Fruit Fly Host Information (CoFFHI), Edition 3.0.
- Steiner, L.F. (1957). Field evaluation of oriental fruit fly insecticides in Hawaii. *J. Econ. Entomol.* **50**, 16–24.
- Xu, L.L., Zhou, C.M., Xiao, Y., Zhang, P.F., Tang, Y., and Xu, Y.J. (2012). Insect oviposition plasticity in response to host availability: the case of the tephritid fruit fly *Bactrocera dorsalis*. *Ecol. Entomol.* **37**, 446–452.
- Dumenil, C., Woud, D., Pinto, F., Alkema, J.T., Jansen, I., Van der Geest, A.M., Roessingh, S., and Billeter, J.C. (2016). Pheromonal cues deposited by mated females convey social information about egg-laying sites in *Drosophila melanogaster*. *J. Chem. Ecol.* **42**, 259–269.
- Zhang, J., Bisch-Knaden, S., Fandino, R.A., Yan, S.W., Obiero, G.F., Grosse-Wilde, E., Hansson, B.S., and Knaden, M. (2019). The olfactory coreceptor IR8a governs larval feces-mediated competition avoidance in a hawkmoth. *Proc. Natl. Acad. Sci. USA* **116**, 21828–21833.
- Hilker, M., and Fatouros, N.E. (2015). Plant responses to insect egg deposition. *Annu. Rev. Entomol.* **60**, 493–515.
- Kellner, R.L.L. (2002). *The Role of Microorganisms for Eggs and Progeny* (Oxford, UK: Blackwell Publishing).
- Mayer, C.J., Vilcinskas, A., and Gross, J. (2008). Pathogen-induced release of plant allomone manipulates vector insect behavior. *J. Chem. Ecol.* **34**, 1518–1522.
- Shikano, I., Rosa, C., Tan, C.W., and Felton, G.W. (2017). Tritrophic interactions: microbe-mediated plant effects on insect herbivores. *Annu. Rev. Phytopathol.* **55**, 313–331.
- Vargas, R.I., Pinero, J.C., and Leblanc, L. (2015). An overview of pest species of *Bactrocera* fruit flies (Diptera: Tephritidae) and the integration of biopesticides with other biological approaches for their management with a focus on the Pacific region. *Insects* **6**, 297–318.