

Inside JEB highlights the key developments in *The Journal of Experimental Biology*. Written by science journalists, the short reports give the inside view of the science in JEB.

FLIES ANTI-TRACK TO EVADE FOUL SMELLS



Flies live in an intensely olfactory world. Tracking mates and homing in on treats, flies are guided by their keen sense of smell. Explaining that flies turn constantly when exploring to locate luscious odours, Sara Wasserman from the University of California Los Angeles, USA, adds that when vinegar flies latch onto an attractive odour trail – such as apple cider vinegar – they stop turning, align their antennae with the steepest gradient of the odour, and track the plume until it guides them to the source. But how would the same insect react when faced with a repulsive odour? Would it turn randomly in a bid to evade the stench or would it latch on to the plume, but pursue it in the opposite direction, anti-tracking it as it became increasingly dilute? ‘We thought they would just avoid the area where the aversive odour was coming from’, recalled Wasserman, so she and her colleagues tested vinegar flies’ responses to an odour that the insects find particularly repulsive: benzaldehyde (p. 2833).

Describing the high-tech flight arena where she and her colleagues, Patrick Lu, Jacob Aptekar and Mark Frye, tested the insect’s aversive behaviour, Wasserman says, ‘We give the flies an odour and look at where they orient.’ Gently gluing a small pin to a fly’s back and attaching it to a frictionless magnetic bearing that allowed the fly to swivel and change direction, Wasserman generated a plume of unpleasant benzaldehyde odour that wafted over the fly and watched the insect’s response. Amazingly, it instantly turned tail and tried to head away from the odour, following the direction of the plume.

‘I ran and got Mark and showed him and we both said, “Let’s see 10 more flies”’, recalls Wasserman, who couldn’t believe the clear-cut response. However, all of the subsequent flies reacted in the same way: the insects were anti-tracking the plume to evade the odour. Could they be using the same mechanism to avoid repellent odours as they use to track attractive scents, but simply switching the direction of their response?

Knowing that the flies found it difficult to position themselves correctly in an attractive plume when the odour receptors on one antenna were blocked with glue – effectively blocking one ‘nostril’ – the team decided to test how flies with a blocked antenna reacted to a jet of repellent benzaldehyde to see whether they were using the same tracking algorithm as the flies without blocked antennae. Dabbing a spot of glue on the third segment of one of the fly’s antennae and positioning it sideways across the odour current, Wasserman saw that the blocked flies were unable to antitrack the repulsive odour – just like the blocked flies in an attractive plume.

Finally, she secured flies to a rigid tether – where she could control the movement of the fly’s visual world in response to their flight patterns to find out how visually alert they were – and found that the odour increased the fly’s alertness, while the visual environment affected their ability to anti-track the repellent plume. ‘This suggests that they utilise the same underlying strategy for tracking an attractive odour and an aversive odour and they just flip the sign somewhere, depending on whether they find it attractive or aversive’, says Wasserman, who is keen to locate the neural switch in order to better understand how flies integrate sensory stimuli.

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Wasserman, S., Lu, P., Aptekar, J. W. and Frye, M. A. (2012). Flies dynamically anti-track, rather than ballistically escape, aversive odor during flight. *J. Exp. Biol.* **215**, 2833-2840.

Kathryn Knight

SANDHOPPERS USE SKYLIGHT GRADIENT TO RECOGNISE SUN

Surrounded by the trappings of modern life, we rarely depend on the sun and moon for guidance, but for sandhoppers scurrying across beaches the correct identification of a celestial body could be a matter of life and death. ‘The ecological problem that sandhoppers have to solve is to stay or return to the damp zone of sand’, says Alberto Ugolini from the Università di Firenze, Italy. Explaining that sandhoppers remain near damp sand for protection from high temperatures and dehydration, he adds, ‘They rely on a wide range of orienting factors such as local cues and general cues, like the sun and moon for correct orientation.’ However, for the minute crustaceans to navigate using celestial objects, they have to be able to successfully distinguish between the sun and the moon. Explaining that sandhoppers are already known to use an internal clock to compensate for the moon’s shifted cycle



relative to the sun, Ugolini and his colleagues Giuditta Galanti and Luca Mercatelli decided to find out whether sandhoppers use the natural gradient of light in the sky to help them distinguish between the sun and the moon (p. 2814).

Collecting the crustaceans from a nearby beach and returning them to the laboratory, the team shifted the animals' body clocks by altering their sunrise to 18:00 h and sunset to 06:00 h. Having allowed the animals to get over their 'jet lag' and adjust to their new time zone, Ugolini and Galanti transported the animals outside on moonlit nights and during the day to test in which direction they oriented. If the crustaceans could correctly distinguish between their view of the sun and the moon, they would align themselves to the correct day- and night-time orientations. However, if the sandhoppers were relying on their internal clock to tell them whether the object in the sky was the sun or the moon, they would mistake the sun for the moon during daylight and align themselves in the direction that they select when viewing the moon. However, the jet-lagged crustaceans coped well. They recognised the sun and positioned themselves correctly, even though their body clock was telling them to expect to see the moon.

Having confirmed that the sandhoppers could distinguish between the sun and the moon, the trio tested the effect of the natural skylight gradient during the day on the crustaceans' ability to identify the sun. Working with Mercatelli, a physicist from the Istituto Nazionale di Ottica CNR, Italy, the team simulated day and night skies in a Plexiglas dome where they could control the light gradient across the artificial sky and the intensity of the sun and moon simulations. Testing the sandhoppers during the day and night, the team simulated skylight gradients in daytime skies with a sun and recorded the sandhoppers'

orientations. As they had expected, the sandhoppers positioned themselves correctly during the day when they saw the daytime sky. And when the team showed the crustaceans a daytime sky during their physiological night, the crustaceans still positioned themselves in their daytime orientation. Finally, the team removed the light simulating the sun and retested the sandhoppers' reactions during the day and night to daytime skylight gradients alone, and the crustaceans still oriented themselves toward their daytime direction, even during the night.

So sandhoppers use the skylight gradient to correctly identify the sun, and Ugolini is now keen to understand the mechanism of this crustacean's internal physiological clock, which allows it to compensate for the shifting cycle of the moon relative to the sun's diurnal clock.

10.1242/jeb.077313

Ugolini, A., Galanti, G. and Mercatelli, L. (2012). The skylight gradient of luminance helps sandhoppers in sun and moon identification. *J. Exp. Biol.* **215**, 2814-2819.

Kathryn Knight

KININ MECHANISM IN HOUSE CRICKET MALPIGHIAN TUBULES

All creatures regulate their ionic balance by excreting water and ions to carefully maintain their delicate osmotic balance. Most insects do this through the Malpighian tubule, which transports ions and fluid from the haemolymph into the lumen of the tubule *via* the principal cells that make up the structure. Geoff Coast from Birkbeck University of London, UK, explains that one class of insect diuretic hormones, diuretic kinins, binds to stellate cells. However, the Malpighian tubules of house crickets lack this cell type, so Coast decided to investigate the function of one diuretic kinin, Achdo-KII, in the transport of ions from the haemolymph across the cricket principal cell and into the tubule lumen (p. 2774).

Measuring Na⁺, K⁺ and Cl⁻ activity (effective concentration) in principal cells before and after the addition of Achdo-KII, Coast also measured the voltage across the principal cell basolateral membrane (the principal cell membrane that faces the haemolymph) and the tubule fluid secretion rate. Coast admits that building the double-

barrelled ion-selective microelectrodes that he used to measure the intracellular ion activity was particularly challenging, and says, 'I spent months struggling with this and was only successful when I switched to a different type of double-barrelled "piggyback glass" on the recommendation of Mike O'Donnell and Juan Ianowski.' Then, after calculating the electrochemical gradients across the basolateral membrane and estimating the gradients for Na⁺ and K⁺ across the apical membrane (the lumen side of the principal cell), Coast constructed a model for ion transport across the cricket Malpighian tubule in the presence and absence of the diuretic hormone Achdo-KII.

Outlining the ion transport model for Malpighian tubule principal cells that have not been stimulated with diuretic hormone, Coast suggests that a Na⁺ gradient drives the transport of 1 Na⁺ and 1 K⁺ ion for every 2 Cl⁻ ions from the haemolymph into the principal cell while Na⁺ and K⁺ ions are transported in turn out of the cell and into the tubule lumen through cation/H⁺ antiporters driven by a proton gradient. Transport is also restricted: Coast explains that the Cl⁻ gradient across the apical membrane in unstimulated principal cells is more than five times greater than the gradient across the basolateral membrane, resulting in low Cl⁻ transport into the tubule lumen, raising the lumen voltage and limiting Na⁺ and K⁺ transport in turn. However, Achdo-KII increases the Cl⁻ conductance across the apical cell membrane into the lumen, resulting in increased Na⁺ and K⁺ transport across the principal cell and into the tubule lumen.

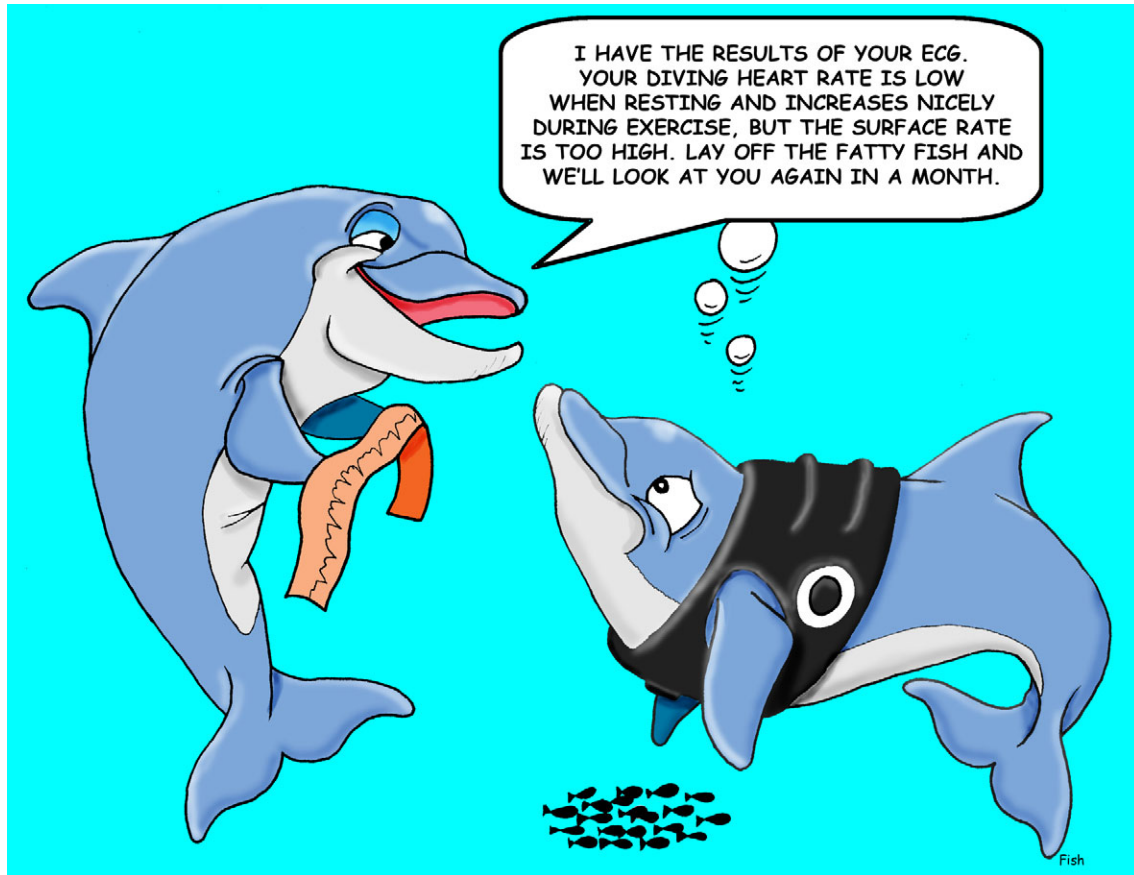
Comparing the cricket Malpighian tubule with the Malpighian tubule of the blood-sucking insect *Rhodnius prolixus*, Coast points out that like the cricket, *R. prolixus* Malpighian tubules also lack stellate cells and that the transport mechanisms in both Malpighian tubules are very similar. He adds, 'This work could readily be expanded upon to investigate the mode of action of another important family of insect diuretic hormones, the corticotrophin releasing factor (CRF)-related neuropeptides.'

10.1242/jeb.077297

Coast, G. M. (2012). Intracellular Na⁺, K⁺ and Cl⁻ activities in *Acheta domesticus* Malpighian tubules and the response to a diuretic kinin neuropeptide. *J. Exp. Biol.* **215**, 2774-2785.

Kathryn Knight

DIVING DOLPHINS RAISE EXERCISE HEART RATE



Submerged for tens of minutes at a time, all diving birds and mammals depart the surface with well-stocked oxygen stores in their blood, muscles and other tissues. But with a finite supply of oxygen in their internal SCUBA tanks, diving animals have to preserve the oxygen they carry in order to return to the surface safely. Terrie Williams and Shawn Noren from the University of California Santa Cruz, USA, explain that diving animals dramatically reduce their heart rate and restrict peripheral blood flow while submerged, and that this reaction is known as the dive response. However, they point out, ‘A paradox exists when marine mammals are foraging underwater because activity should promote an elevation in heart rate to support increased metabolic demands.’ So could diving mammals increase their heart rates during exercise despite the pressure to conserve oxygen supplies (p. 2735)?

Teaming up with Traci Kendall and Veronica Cuccurullo, Williams travelled to the Bahamas to put trained adult bottlenose dolphins through their metabolic paces. Fitting the animals with custom-made neoprene vests carrying electrocardiograph monitors, the team filmed the diving dolphins as they performed a range of underwater activities – ranging from sitting still or bobbing their heads up and down to swimming at their speed of choice – while measuring the animals’ heart rates.

Analysing the heart traces, Noren confirmed that the resting dolphins dropped their heart rates dramatically from 105 beats min^{-1} at the surface to 40 beats min^{-1} while submerged. However, as the team asked the dolphins to perform more activities, the animals’ heart rates increased to 56 beats min^{-1} while head

bobbing to 71 beats min^{-1} when swimming. The animals also raised their heart rates even further in anticipation of returning to the surface during their ascents.

So, diving dolphins can moderate their dive response to accommodate their activity levels and the team says, ‘Alterations in blood flow throughout submergence theoretically facilitates more effective unloading of endogenous oxygen stored by enabling the parallel depletion of the blood and muscle oxygen reserves.’

10.1242/jeb.077305

Noren, S. R., Kendall, T., Cuccurullo, V. and Williams, T. M. (2012). The dive response redefined: underwater behavior influences cardiac variability in freely diving dolphins. *J. Exp. Biol.* **215**, 2735-2741.

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