

Inside JEB is a twice monthly feature, which 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.

Inside JEB

BATS MODULATE CALLS FOR INCREASED ACCURACY



Embarking on twilight foraging excursions, bats have only their ears to guide them through cluttered environments. Uttering high-pitched cries and listening for their reflections, bats construct a sonic view of their world. But no two echolocation calls are the same. Marc Holderied, from the University of Erlangen, Germany, explains that each call is optimised for a different circumstance. Cries ranging over a narrow frequency-range are better suited for long-range echolocation, while cries swooping over a wide frequency-range function best for short-range detection. But no one knew whether bats tailored their calls to improve their navigational precision. Holderied and his colleague Otto von Helversen decided to record the positions and signal structures of echolocating bats to see if the bats fine-tuned their calls in response to the obstacles they encounter (p. 1816).

Carefully rigging up eight microphones close to the bat's path, Holderied recorded 22 individuals' echolocating cries as the tiny aviators departed their farmhouse colony and followed a hedge to their nearby hunting grounds. But Holderied also wanted to relate the bats' calls to their surroundings, so he surveyed the bats' guiding hedge with a theodolite and laser to reconstruct the hedge's structure.

Teaming up with Gareth Jones at the University of Bristol, UK, Holderied calculated the position of each bat relative to the hedge when it squeaked, and the cry's duration to see whether the animal's call length changed as they tracked the hedge. But when he analysed each call's duration, the animal's proximity to the hedge seemed to have no effect. The bats weren't moderating their call length to prevent the outgoing call interfering with the incoming reflection, as he had thought.

However, when Holderied analysed each call's bandwidth as the bats progressed along the hedge, there was a strong correlation; when the bats neared the hedge,

they increased the bandwidth of their cries. Holderied explains that increasing the bandwidth allows the bats to estimate the distance to near-by objects more accurately.

Curious to know if the bats modulated their cries in a systematic way as they followed the hedge's contours, the team decided to calculate each cry's 'distance of focus'. Holderied explains that the distance of focus is the distance where the squeaking bat can accurately estimate the position of obstacles; there are always errors in the flying bat's distance measurements if the obstacle is nearer to, or further away, than the distance of focus. Calculating each cry's distance of focus from the squeak's bandwidth and duration, and plotting it as a hemisphere centred on and in front of the bat, Holderied realised that the bats continually adjusted their calls so that the distance of focus skirted the hedge's foliage as they progressed along its length. The bats were continually adapting their calls to follow the hedge with maximum precision.

Holderied suspects that echolocating bats are able to select calls with distances of focus that roughly match the location of an object as it looms, freeing the bats from complex echolocation calculations and improving their in-flight precision.

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Holderied, M. W., Jones, G. and von Helversen, O. (2006). Flight and echolocation behaviour of whiskered bats commuting along a hedgerow: range-dependent sonar signal design, Doppler tolerance and evidence for 'acoustic focussing'. *J. Exp. Biol.* **209**, 1816-1826.

DIMM REGULATES NEUROPEPTIDE LEVELS

There are few molecules in biology as powerful as neuropeptides. Regulating a host of physiological functions from growth and reproduction to sleep and circadian rhythms, neuropeptides are essential for many aspects of life. Randall Hewes from the University of Oklahoma explains that neuropeptide expression levels vary hugely in response to physiological stimuli, and that the expression of these neural signalling molecules is tightly regulated. One of the few proteins that is known to regulate neuropeptide levels in *Drosophila* is the DIMM transcription factor, which is expressed widely in neural tissue. But which neuropeptides are regulated by DIMM and how the transcription factor functions wasn't clear. Hewes and Sebastian Gauthier began comparing mutant *Drosophila* that lacked a functional *dimm* gene with natural flies to see which neuropeptides were regulated by the transcription factor (p. 1803).

The team decided to compare the cellular levels of mRNA for 16 neuropeptide and associated protein genes in mutant fly larvae, which lacked a functional *dimm* gene, with the mRNA levels of the same neuropeptides in natural fly larvae, which retained the intact transcription factor gene. But the team faced a complication. Hewes explains that all of the *dimm* mutant flies also carried a mutation in another transcription factor gene, *cryptocephal*, due to difficulties controlling the exact location of the mutation in *Drosophila*. So the team had to make sure that they also monitored the 16 genes' mRNA levels in fly larvae where *cryptocephal* alone was mutated to make sure that any differences between the mutant and natural flies were due to the *dimm* gene.

Quantifying the mRNA level differences between the natural and mutated fly larvae, Gauthier and Hewes clearly saw that the mRNA from three neuropeptide genes fell dramatically in the mutant animals: *Dromyosuppressin*, *FMFRamide-related* and *Leucokinin* were all transcriptionally regulated by DIMM. The team suspects that DIMM also regulates the protein levels of other neuropeptide genes through other cellular mechanisms.

However, the team was in for a shock when they looked at the mRNA levels in their control flies. All but one of the monitored genes behaved exactly like the genes in the natural fly larvae, but when they measured the level of Ecdysis triggering hormone (ETH) in the *cryptocephal* mutant flies, the hormone's level was severely reduced. *Cryptocephal* controls ETH levels. They had inadvertently discovered one of the genes that *cryptocephal* targets.

Hewes explains that the *cryptocephal* mutation was first identified in 1942, when flies lacking the gene were found to fail to complete ecdysis. Now that Hewes and Gauthier have found this critical link between *cryptocephal* and the hormone that orchestrates ecdysis, they would like to know exactly how *cryptocephal* regulates ETH levels.

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Gauthier, S. A. and Hewes, R. S. (2006). Transcriptional regulation of neuropeptide and

peptide hormone expression by the *Drosophila dimmed* and *cryptocephal* genes. *J. Exp. Biol.* **209**, 1803-1815.

FIRST 'SPECIFIC' INHIBITORY NEURON IN INSECTS



Picture by Harald Wolf

Peter Bräunig, Michael Schmäh and Harald Wolf are fascinated by the neurophysiology of locusts, but each from a slightly different perspective: Wolf is intrigued by the role of inhibitory neurones in neuromuscular control, Bräunig by the neuroanatomy of the insect's head and thorax, and Schmäh by the insect's segmental organisation. Wolf explains that the three came together as a team when Bräunig noticed that a region of the insect's thorax was supplied by more neurones than expected. Their curiosity aroused, the team began characterising the insect's neurophysiology and were surprised to find that unlike all other arthropod inhibitory neurones, which function on several muscles simultaneously, one of the thorax's inhibitory neurones was 'specific', functioning on one muscle alone (p. 1827).

Wolf explains that unlike large animals, which selectively activate fast and slow fibres through a myriad individual motoneurons, muscle function in smaller arthropods is controlled by as few as two or three motoneurons activating both slow and fast muscle fibres. According to Wolf, slow muscle fibres tend to be activated first, while fast muscle fibres are activated only at higher neurone discharge rates during an excitatory signal. So how do arthropods move fast when the muscle fibres that are initially activated are designed for slow endurance performance? Wolf explains that arthropod muscles are innervated with inhibitory neurones that act only on slow

muscle fibres, effectively switching off the slow muscle fibres when the creatures need to get a move on. Wolf adds that almost all inhibitory neurones act on several muscles simultaneously, which he explains makes sense; if a crab wants to walk fast, it might as well inhibit all the slow muscle fibres in its leg muscles with a single neuron. However, only two inhibitory neurones, found in decapod crustacean legs, are known to function 'specifically' on single muscles, allowing the crustaceans to independently activate two muscles that are excited by a single neuron. Curious to know which neurones in the locust's thorax were inhibitory, the team traced the delicate neural tissue through a segment of the insect's thorax.

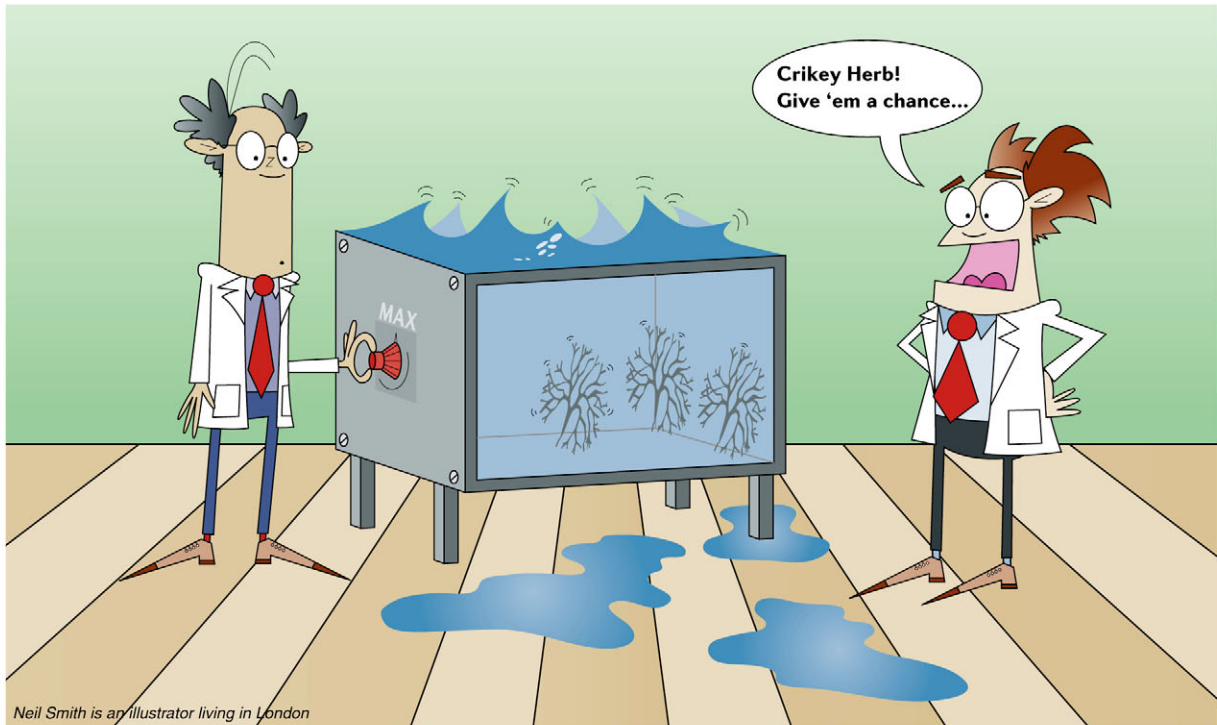
The team washed thorax nerves with antisera that stained neurones producing the inhibitory neurotransmitter, GABA. Surprisingly, three of the neurones were inhibitory, even though most body segments are inhibited by only two. Curious to know which muscles were innervated by the inhibitory neurones, the team infused the insect's nervous tissue with cobalt and nickel ions, to track the neurones's course. Sure enough, two of the neurones served several muscles, but when the team traced the third, it only inhibited one muscle, known as M60. The team confirmed their unexpected finding by measuring electrical activity on the three neurones, and correlated their activity with signals in the muscle tissue.

So why is the inhibitory neuron that innervates M60 'specific'? Wolf suspects that there are several possible explanations. Either the neuron was previously connected to another muscle and is in the process of losing that connection, or the segment with three inhibitory neurones is an evolutionary snapshot, linking the locust to early dragonflies. Wolf explains that each modern dragonfly segment is innervated by three inhibitory neurones, and the locust could prove to be the missing link between the neurophysiology of early dragonflies and modern insects.

10.1242/jeb.02265

Bräunig, P., Schmäh, M. and Wolf, H. (2006). Common and specific inhibitory motor neurones innervate the intersegmental muscles in the locust thorax. *J. Exp. Biol.* **209**, 1827-1836.

CHANGING SHAPE TO GO WITH THE FLOW



Clinging to the seashore, life is a constant tug-of-war. Continually wrenched about, seashore dwellers have a range of strategies for withstanding the constant battering. Some creatures have armoured themselves to withstand the pounding, while others have chosen to go with the flow, changing shape and bending with the current. Michael Boller explains that although it is well known that seaweeds continually change shape in the surf, it wasn't clear which mechanisms protect macroalgae's delicate fronds. To find out how seaweeds adjust to the rigours of sea life, Boller and Emily Carrington

measured the forces exerted on *Chondrus crispus* in currents ranging from 0-2 m s⁻¹ (p. 1894). Filming the seaweed's movements, the pair found that at lower speeds, *Chondrus crispus* simply deflected to align with the direction of flow. However, as the flow increased the seaweed's crown became more compacted, reducing the drag experienced by the macroalage.

Having quantified how the macroalgae's drag coefficient and frontal area decrease as the current picks up, the scientists developed a mathematical model to

calculate the drag experienced by flexible structures in water, which they suggest 'improves our ability to predict drag at higher, ecologically relevant water velocities'.

10.1242/jeb.02264

Boller, M. L. and Carrington, E. (2006). The hydrodynamic effects of shape and size change during reconfiguration of a flexible macroalga. *J. Exp. Biol.* **209**, 1894-1903.

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