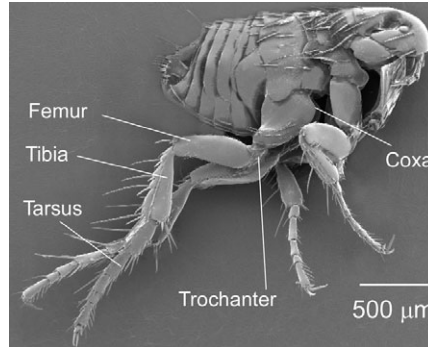


Inside JEB highlights the key developments in *The Journal of Experimental Biology*. Written by science journalists, the short reports give the inside view of the science in JEB.

Inside JEB

MYSTERY OF FLEA JUMP RESOLVED: TARSUS PUSH OFF



If you thought that we know everything about how the flea jumps, think again. In 1967, Henry Bennet-Clark discovered that fleas store the energy needed to catapult themselves into the air in an elastic pad made of resilin. However, in the intervening years debate raged about exactly how fleas harness this explosive energy. Bennet-Clark and Miriam Rothschild came up with competing hypotheses, but neither had access to the high speed recording equipment that could resolve the problem. Turn the clock forward to Malcolm Burrows' Cambridge lab in 2010. 'We were always very puzzled by this debate because we'd read the papers and both Henry and Miriam put a lot of evidence for their hypotheses in place and their data were consistent with each other but we couldn't understand why the debate hadn't been settled,' says Burrows' postdoc, Gregory Sutton. He adds, 'We had a serendipitous set of hedgehog fleas show up so we figured we'd take a crack at it and try to answer the question' (p. 836).

'We were concerned about how difficult it would be to make the movies because we are used to filming locusts, which are much bigger than fleas,' admits Sutton, but he and Burrows realised that the fleas stayed perfectly still in the dark and only jumped when the lights went on. Focusing the camera on the stationary insects in low light, the duo successfully filmed 51 jumps from 10 animals; and this was when they got their first clue as to how the insects jump.

In the majority of the jumps, two parts of the flea's complicated leg – the tarsus and trochanter – were in contact with the ground for the push off, but in 10% of the jumps, only the tarsus touched the ground. Sutton explains that Rothschild had suggested that fleas push off with the trochanter, but if 10% of the jumps didn't use the trochanter was it really necessary, or were the fleas using two mechanisms to get airborne?

Burrows and Sutton needed more evidence. Analysing the movies, the duo could see

that the insects continued accelerating during take-off, even when the trochanter was no longer pushing down. And the insects that jumped without using the trochanter accelerated in exactly the same way as the insects that jumped using the trochanter and tarsus. Also, when Burrows and Sutton looked at the flea's leg with scanning electron microscopy, the tibia and tarsus were equipped with gripping claws, but the trochanter was completely smooth. Sutton and Burrows suspected that the insects push down through the tibia onto the tarsus, as Bennet-Clark suggested, but the team needed one more line of evidence to clinch the argument: a mathematical model that could reproduce the flea's trajectory.

'I looked at the simplest way to represent both models,' explains Sutton. Building Rothschild's model as a simple mass attached to a spring pushing down through the trochanter and Bennet-Clark's model as a spring transmitting the spring's force through a system of levers pushing on the tarsus, Sutton generated the equations that could be used to calculate the insect's trajectory. Then he compared the results from his calculations with the movies to see how well they agreed.

Both models correctly predicted the insect's take-off velocity at 1.35 m s^{-1} , but then the Rothschild model began to go wrong. It predicted that the insect's acceleration peaked at a colossal $22,000 \text{ m s}^{-2}$ (2200 g), whereas the acceleration of the insects in the movies only peaked at 1500 m s^{-2} (150 g). However, Sutton's calculations based on the Bennet-Clark lever model worked perfectly, accurately predicting the insect's trajectory and acceleration pattern.

So Sutton and Burrows have finally settled the argument and resolved how fleas jump. The insects transmit the force from the resilin spring in the thorax through leg segments acting as levers to push down on the tarsus and launch the 0.7 mg animals at speeds as high as 1.9 m s^{-1} .

10.1242/jeb.056408

Sutton, G. P. and Burrows, M. (2011). Biomechanics of jumping in the flea. *J. Exp. Biol.* **214**, 836-847.

DO POND SNAILS SLEEP?

Sleep is a precious commodity. Anyone who suffers from insomnia knows how crippling sleep loss is. Richard Stephenson and Vern Lewis from the University of Toronto, Canada, explain that sleep is thought to play a pivotal role in a range of biological processes, including memory formation, but many questions still remain



Vern Lewis

unanswered. For example, we don't even know how much sleep we need. According to Stephenson and Lewis, molluscs – such as the sea hare and great pond snail (*Lymnaea stagnalis*) – have taught us a great deal about the neural basis of memory formation, but could they teach us about the mechanisms of sleep: especially as it wasn't clear whether they do it. Stephenson and Lewis decided to find out whether snails sleep (p. 747).

'There is no single characteristic that unequivocally defines the sleep state,' say Stephenson and Lewis, 'instead several criteria are used collectively.' Describing how sleeping animals are usually unresponsive, hard to wake and settle in a characteristic position, the duo monitored the behaviour of a tank full of pond snails, cataloguing the molluscs' activities to find out whether they did anything that looked like sleep.

Analysing the snails' behaviours, Stephenson and Lewis clearly saw them attached to solid surfaces and inactive for periods of tens of minutes. And the snails looked relaxed like other sleeping species: their shells hung away from the body while attached to the side of the tank, the foot looked symmetrical and relaxed, and their tentacles were only partially extended.

Having identified a sleep-like resting state, the duo tested the snails' responses: they tapped the molluscs and stimulated their appetites when active and apparently sleeping. The scientists found that the resting snails were much slower to react than active snails, taking over twice as long to retract into their shells when poked and seven times as long to respond when their appetite was stimulated. All in all, the snails certainly seemed to be sleeping, but was their sleep rhythmic like ours and would it be affected by day length?

The team monitored the behaviour of 8 snails over 79 days as they varied the animals' light exposure. Instead of regulating their sleep over a 24 h period, the snails clustered sleep bouts in a pattern that cycled every 2–3 days. Also, they did not

seem to suffer 'sleep rebound' – when we make up for lost sleep.

Despite the differences between mammalian and pond snail resting behaviours, Stephenson and Lewis believe that great pond snails do sleep. They say, 'We suggest that *Lymnaea stagnalis*, by virtue of its anatomical simplicity and neurophysiological tractability, may prove useful in the investigation of cellular mechanisms of sleep regulation and sleep function'.

10.1242/jeb.056390

Stephenson, R. and Lewis, V. (2011). Behavioural evidence for a sleep-like quiescent state in a pulmonate mollusc, *Lymnaea stagnalis* (Linnaeus). *J. Exp. Biol.* **214**, 747–756.

BRAIN CELLS BORN WHEN ELECTRIC FISH BREED



Matias Galeano

Until the 1970s, researchers believed that adult animals' brains were pretty much 'fixed'. But in recent decades, neuroscientists have discovered just how flexible adult brains are. Exposure to complex surroundings can even boost the birth of new brain cells – at least in birds and mammals. Kent Dunlap, a biologist at Trinity College, Hartford, USA, wondered whether the same would be true for fish (p. 794).

To find out, Dunlap decided to take a closer look at electric knifefish brains. Studying electric fish has two advantages, he says. First, electric fish navigate and communicate using specific brain regions that generate and process electric signals; that is, certain brain regions help fish cope with the challenges posed by their physical and social environments. Second, fish take on the temperature of their surroundings – allowing Dunlap to examine whether seasonal changes in environmental temperature influence brain cell birth rates.

Teaming up with Ana Silva at the Instituto de Investigaciones Biológicas Clemente Estable in Uruguay and Mike Chung at Trinity College, Dunlap set out to see how three different environments – natural,

semi-natural and isolated – affect brain cell birth rates across different brain regions in electric knifefish. The team first headed to lake Laguna Lavelle in Uruguay to study fish in their natural home during the breeding season. To label newborn brain cells, they caught wild fish and injected them with bromodeoxyuridine (BrdU), an analogue of one of the building blocks of DNA that is incorporated into dividing cells. Then they froze the fish brains, sliced them into thin sections and used anti-BrdU fluorescent antibodies to make the newborn brain cells glow, so that they could count how many there were in different brain sections. To compare brain cell birth rates in wild and captive fish, the team also took fish back to the lab. They housed some fish in groups in small paddling pools and others alone in aquaria. To see how fish brains respond to changing seasons, the team repeated the brain cell labelling process for all three fish populations in the non-breeding season a few months later.

'After just 2 days of looking at brain sections, it was clear that seasonality had a huge effect,' Dunlap recalls. During the breeding season, Silva explains, fish had 3–7 times more newborn brain cells than during the non-breeding season. 'This suggests that warm temperatures and long day lengths not only trigger reproduction in this temperate zone species but may also increase brain cell proliferation,' says Silva.

But did differences in the physical and social environment also affect brain cell birth rates? Sure enough, just as in birds and mammals, more brain cells are born in fish living in a complex environment; the team found that lake-dwelling fish had higher brain cell birth rates across all brain regions than captive fish. When the team examined the brain sections more closely, they saw that socially housed fish had more newborn brain cells than lonely fish – but only during the breeding season, and only in brain regions involved in electrocommunication. In other words, when they need to woo prospective partners, electric fish pump up brain cell production in those brain regions that boost social signalling prowess. 'Small changes like living socially have effects on specific brain regions, while big changes like seasonality cause global changes across the whole brain,' Dunlap concludes and adds, 'when the global seasonal effect combines with the specific social effect, brains produce cells especially rapidly.'

10.1242/jeb.056416

Dunlap, K. D., Silva, A. C. and Chung, M. (2011). Environmental complexity, seasonality and brain cell proliferation in a weakly electric fish, *Brachyhyponomus gauderio*. *J. Exp. Biol.* **214**, 794–805.

Yfke Hager

MASS REEF SPAWNING COULD BE TRIGGERED BY TWILIGHT



Reproduction can be a haphazard event, especially when you simply cast your gametes into the sea and hope for the best. However, some organisms in coral reefs have improved their odds by synchronising when they spawn. These mass spawning events on reefs can last for as little as 20 min and only occur during twilight for a few nights each year. So, how do animals that lack even the simplest of nervous systems coordinate such sophisticated behaviour? Alison Sweeney and her colleagues from the University of California, Santa Barbara, and Duke University wondered whether mass spawning events might be synchronised by fluctuations in the twilight spectrum. They explain that the spectrum of twilight is deep blue before the moon rises, but after moonrise the spectrum becomes redder. As the moon is already in the sky at sunset during the first half of a lunar month, the twilight spectrum is always red shifted, but at full moon (when the moon is just below

the horizon at sunset) there is a brief period when the spectrum of skylight is deep twilight blue before the moon rises. Sweeney and her colleagues realised that corals and other reef residents could use this brief period of pure twilight to synchronise spawning, but only if the spectrum of light in the ocean followed the same pattern as skylight (p. 770).

Measuring the spectrum of light in the ocean above a coral reef in the US Virgin Islands over a 6 day period around full moon, the team found that the twilight spectrum shifted significantly depending on whether or not the moon had risen. At full moon, the twilight spectrum was deep blue just after sunset but gained red wavelengths as soon as the moon rose. Also, the length of the blue twilight period increased on subsequent evenings as the moon rose later each day. While recording the light spectrum, the team also monitored elkhorn coral colonies for spawning events and

found that the corals spawned simultaneously between 21:30h and 21:50h on the third and fourth nights after the full moon.

Of course, this does not confirm that corals use shifts in the twilight spectrum to synchronise spawning – at present there is only a correlation. However, Sweeney and her colleagues point out that corals are capable of detecting changes in light quality and synchronise successfully even on overcast days, so they are keen to test the effects of skylight spectra on spawning events to find out whether twilight synchronises mass spawning.

10.1242/jeb.056382

Sweeney, A. M., Boch, C. A., Johnsen, S. and Morse, D. E. (2011). Twilight spectral dynamics and the coral reef invertebrate spawning response. *J. Exp. Biol.* **214**, 770-777.

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