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Inside JEB

METH(AMPHETAMINE) MAY STOP SNAILS FROM FORGETTING


Ken Lukowiak

Crystal meth (methamphetamine) is a highly addictive drug that seduces victims by increasing self-esteem and sexual pleasure, and inducing euphoria. But once hooked, addicts find the habit hard to break. Barbara Sorg from Washington State University, USA, explains that amphetamines enhance memory. ‘In addiction we talk about the “drug memory” as a “pathological memory”. It is so potent as to not be easily forgotten,’ she explains. As memory plays an important role in addiction, Sorg wondered whether it might be possible to find out more about the effects of meth on memory by looking at the effect it has on a humble mollusc: the pond snail *Lymnaea stagnalis* (p. 2055).

Lymnaea hold memories about when to breathe through their breathing tubes (pneumostomes) in a three neuron network, which is much simpler than the colossal circuits that hold our memories. Ken Lukowiak from University of Calgary, Canada, has been working on the mechanisms of memory formation in these snails for most of his career, so he and Sorg decided to team up to find out whether a dose of meth could improve the snails’ memories in the way it does human memories.

First Sorg and her students had to discover whether a dose of meth could affect the snails’ breathing behaviour. According to Lukowiak, the snails breathe through their skins when oxygen levels are high, but when oxygen levels drop the snails extend their pneumostomes above the water’s surface to supplement the supply. As the drug easily crosses the snail’s skin, the team immersed the snails in de-oxygenated pond water spiked with meth, and watched to see how it affected their breathing. The snails stopped raising their pnemostomes at 1 and 3.3 μmol l^{-1} meth, so having found a dose that altered the snail’s behaviour, the team began testing its effects on the mollusc’s long term memory.

The team trained the snails to remember to keep their pneumostomes closed when oxygen levels were low by poking them with a stick every time they tried to open their pneumostomes. Giving the snails two training sessions separated by an hour, the team knew that the molluscs would hold the memory for over 24 h, but what would happen if they trained the snails in meth-laced water?

Testing the snails in de-oxygenated pond water 24 h later, the team were surprised to see that the snails seemed to have no recollection of their training, popping their pneumostomes above the water’s surface. Maybe meth did not affect the snails’ memories. But then Lukowiak remembered: ‘If you put snails in a novel context even though they have memory they respond as if they don’t have memory,’ he says. Without meth in the water, the snails were ignoring their memory. However, when the team reintroduced meth to the test water, the snails suddenly remembered to keep their pneumostomes closed. This could explain why it’s so hard for human addicts to kick the habit when returning to old haunts that trigger the addiction memory.

Next the team wondered whether meth could improve the snails’ memories. First they immersed the snails in meth-laced pond water, then they moved them into regular de-oxygented pond water and gave them a training session that the snails should only recall for a few hours. In theory the snails should have forgotten their training 24 h later, but would the meth improve the snails’ memories so they remembered to keep their pneomostomes closed a day later? It did. A dose of meth prior to training had improved the snails’ memories, allowing them to recall a lesson that they should have already forgotten. And when the team tested whether they could mask the meth memory with another memory, they found that the meth memory was much stronger and harder to mask.

So memories formed under the influence of meth seem to be harder to forget, possibly because the drug disrupts the mechanisms for forgetting, and could help us to understand how amphetamines enhance memory in humans.

10.1242/jeb.046664

Kennedy, C. D., Houmes, S. W., Wyrick, K. L., Kammerzell, S. M., Lukowiak, K. and Sorg, B. A. (2010). Methamphetamine enhances memory of operantly conditioned respiratory behavior in the snail *Lymnaea stagnalis*. *J. Exp. Biol.* **213**, 2055-2065.

AMPK ACTIVATOR ACTIVATES HIBERNATOR’S APPETITE

Some of us may put a few inches on over the course of winter, but not hibernating animals. They gain fat during the late summer ready for fuel when they stop feeding and begin hibernating. But what controls this dramatic


Greg Florant

switch from gorging to fasting? Gregory Florant from Colorado State University and his colleagues from the University of Arizona – Phoenix, USA, knew that AMP-activated protein kinase (AMPK) acts as an intracellular energy sensor and regulates food intake, so the team decided to test whether it may also play a role in regulating hibernator's eating habits (p. 2031).

Collecting nine yellow-bellied marmots in Colorado, the team kept the animals well supplied with food in a temperature regulated room as they simulated the onset of winter. By January all of the marmots had been fasting and hibernating for 3 months, despite being well supplied with food, so the team infused a compound that activates AMPK into the animals' brains to see what effect it had on them.

Amazingly the animals that received the AMPK activator began eating and some even gained weight, while the animals that were infused with a simple saline solution continued fasting and lost weight. Admittedly the team did have to relocate the marmots to a warmer room as they received the AMPK activator, but the saline treated animals also lowered their body temperatures and continued hibernating while the AMPK activated animals became active and stayed warm.

So a dose of an AMPK activator is able to switch on a hibernator's urge to eat, probably by activating the intracellular energy sensor AMPK, and the team are now keen to identify the neural pathways that regulate a hibernator's appetite.

10.1242/jeb.046672

Florant, G. L., Fenn, A. M., Healy, J. E., Wilkerson, G. K. and Handa, R. J. (2010). To eat or not to eat: the effect of AICAR on food intake regulation in yellow-bellied marmots (*Marmota flaviventris*). *J. Exp. Biol.* **213**, 2031-2037.

SQUID FINS PRODUCE LIFT, THRUST AND STABILITY

Squid are remarkably versatile swimmers. While most fish find it difficult to reverse, squid are equally happy going backward and forward, swimming tail first at high speeds but arms first when hovering and swimming slowly to survey the area. Although squid generate most of their thrust at high speeds by squeezing jets of water out of the mantel, they are equipped with a versatile pair of mantle fins that ripple and flap at lower speeds. But it wasn't clear exactly how the fins contributed to the animal's agility. Were the fins simply acting as stabilisers, or could they generate thrust to propel the animals? Having previously investigated the way that squid jet around at higher speeds, William Stewart and Ian Bartol from Old Dominion University, USA, and Paul Krueger from Southern Methodist University, USA, decided to find out more about the way the squid use their fins while swimming (p. 2009).

After successfully catching the elusive animals, Stewart and Bartol rushed back to the lab ready to put them through their paces in a flume. Adding microscopic beads to the water and shining a plane of laser light on the tip of one of the squid's fins to reveal the water's motion the duo filmed the squid as they swam arms first (fins at the back) and tail first (fins at the front) at speeds ranging from 2 cm s^{-1} up to 23 cm s^{-1}. Having collected the data, the trio spent another 6 months analysing it to find out how the squid use their fins and were pleased to see the tell-tale vortices that they had hoped to see spinning off the fins in four distinct modes when the squid swam tail first. However, the animals only used two of the four modes while swimming arms first.

When swimming tail first in the first mode, the squid flapped their fins up and down, but only exerted force on the water during the down-stroke. 'We did not measure any detectible vorticity associated with the up-stroke,' says Stewart and adds that instead of generating thrust the fins mostly produced lift to hold the squid's vertical position in the water column. When looking at the second mode, the team saw that instead of flapping the fins, the squid sent an S-shaped ripple along the fin edge, resulting in a chain of linked vorticity producing weak upward jets and stronger downward jets of water giving rise to a net lift force on the animal's body.


Ian Bartol

Analysing the third and fourth modes, the team realised that instead of generating lift alone, they both produced thrust to propel the squid forward. In the third mode the squid returned to flapping their fins, but the fin beat was relatively leisurely, producing independent vortices at the end of each up- and down-stroke, while in the fourth mode, the up-stroke followed rapidly after the down-stroke so that the shed vortices became linked in pairs.

Turning their attention to the squid's 'arms first' swimming style, the team found that the animal's fins also produced the second and third vorticity patterns, but this time neither pattern generated significant thrust – they both produced lift and often drag.

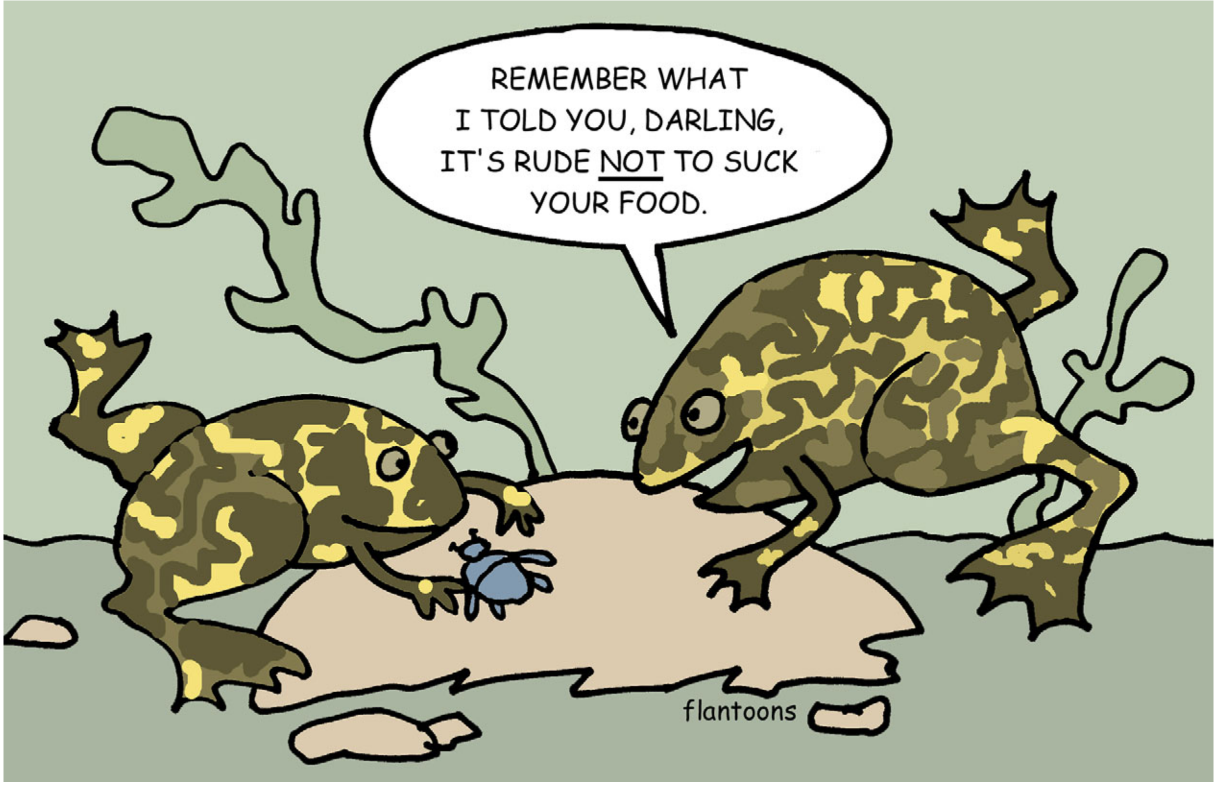
Stewart admits that he was surprised that the squid can produce two of the same vorticity patterns regardless of whether they swim arms or tail first. He explains that instead of being stiffened by rays the fins are muscular hydrostatic systems, and the squid's ability to produce the same vorticity patterns when swimming in opposite directions reflects the fin's versatility.

So squid can use their fins to generate lift and provide stability, but they can also use them to generate thrust to supplement their mantel jets, thanks to their versatile muscular structure.

10.1242/jeb.046649

Stewart, W. J., Bartol, I. K. and Krueger, P. S. (2010). Hydrodynamic fin function of brief squid, *Lolliguncula brevis*. *J. Exp. Biol.* **213**, 2009-2024.

PIPID FROGS SUCK



Catching dinner in water isn't easy. If you lunge forward you'll most likely push the food out of range on your bow wave. Carrie Carreño and Kiisa Nishikawa explain that pipids, an entirely aquatic species of frog, cannot feed like other frogs because they lack a tongue, so how do they do it? One possibility is that they slurp food in by suction. However, it wasn't clear whether some or all pipids feed by suction. Some observers had found that some pipids appear to suck while others sweep food into their mouths with their forelimbs. The duo decided to film four pipid species and measure the pressure in their mouths as they fed to lay the controversy to rest (p. 2001).

Filming Surinam toads, African clawed frogs, dwarf African clawed frogs and Merlin's frogs as they fed, the duo could see that the food began moving before the frogs were close enough to bite. The animals must be sucking it in. And when Carreño and Nishikawa analysed the pressure profiles in the animal's mouths, they found that the pressure did drop and they could suck morsels into their mouths. In addition the African clawed frogs and Surinam toads usually swept their forelimbs in front of them to sweep the morsels towards their mouths.

So all four pipid species can, and do, suck food into their mouths, explaining how pipids have overcome the loss of their tongues when feeding.

10.1242/jeb.046656

Carreño, C. A. and Nishikawa, K. C. (2010). Aquatic feeding in pipid frogs: the use of suction for prey capture. *J. Exp. Biol.* **213**, 2001-2008.

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