

INSIDE JEB

Thermal imaging and vision are equally important for hunting pit vipers



A short-tailed pit viper (*Gloydius brevicaudus*). Photo credit: Qin Chen.

Hunting by day is a doddle when equipped with a sharp pair of eyes, but when the conditions are dim, even the keenest sight isn't going to help, which is where thermal imaging can come in handy. Pit vipers are equipped with specialised infrared-sensing pit organs that provide a thermal perspective of the serpent's surroundings, which can help the predator to pick off warm-blooded animals. Intrigued by the reptile's twin visual systems, Qin Chen, from the Chengdu Institute of Biology, China, explains that it was not clear how much significance the vipers attached to each system. 'How the snakes combine the two into a picture has always fascinated us', says Chen. Knowing that vipers that are equipped with tape blinds can strike successfully at warm and cold objects when viewed with their infrared sensor against an intermediate temperature background, Chen and his colleagues Guangzhan Fang and Yezhong Tang wondered how much of an emphasis the snakes put on their thermal view when the contrast between the warmth of their victim and its surroundings varies.

As Qin Chen had been bitten by a large pit viper (*Protobothrops jerdonii*) previously, the team chose to work with the smaller short-tailed pit viper (*Gloydius brevicaudus*), which is more docile and an adept hunter in bright and dim conditions. After collecting the vipers and transporting them back to Chengdu, Chen and Yang Liu built a drum-shaped arena lined with a heating film so that they could warm one side of the chamber while the other half remained at room temperature (26°C).

The duo then measured the surface temperature of the mice (33°C) that would be pitted against the vipers, and set one half of the arena at that temperature (so that the mouse would blend in) while holding the other at 26°C (which the mouse should stand out brightly from) before filming individual vipers as they attacked the rodent either with or without the benefit of their eyes and thermal pit sensors.

Guided by both their eyes and infrared pits, the vipers successfully landed all nine of their attacks against the cooler background, although they missed two out of the 12 attacks against the 33°C walls. In contrast, when the vipers had to strike with only their pit organs for guidance, their success rate was significantly higher against the 26°C wall – landing 14 successful strikes – compared with the four that they managed to land as the mice scampered across the 33°C background, only two of which succeeded. However, when the snakes had to depend on vision alone, they still missed four of their 10 strikes against the cool cylinder wall, while only landing eight of the nine strikes at mice in front of the low-contrast 33°C background.

Chen and Liu then repeated the experiments, but this time they heated the warmer half up to 40°C while the cooler half remained at 26°C, to find out how the contrast of the rodent's body temperature against the background affected the snake's success. The snakes were 100% successful against the cooler background when guided by their thermal pits alone, although they only landed 55% of the strikes when the mice appeared as a dim thermal shape against the 40°C background.

'Snakes prefer to target prey in front of backgrounds with positive thermal contrasts when only their pits are active', says the team, and they suggest that the vipers are equally dependent on their thermal and visual senses as their success rate fell when the snakes were only able to use their eyes.

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Chen, Q., Liu, Y., Brauth, S. E., Fang, G. and Tang, Y. (2017). The thermal background determines how the infrared and visual

systems interact in pit vipers. *J. Exp. Biol.* **220**, 3103-3109.

Kathryn Knight

Endocannabinoids disrupt stressed snails' memories



Lymnaea stagnalis. Photo credit: Wikimedia.

Life has never been easy, and the physical mechanisms that were evolved by the earliest life forms for protection are fundamental to survival even today. However, fast-forward to our 24/7 news-saturated millennial culture and those stress systems are saturating. 'We all know that when we are under stress we perform differently', says Ken Lukowiak, from the University of Calgary, Canada, who is investigating the impact of stress on memory. However, instead of trying to untangle the complex networks that underpin human memory, Lukowiak and his colleagues focus on the memory of a simpler creature, pond snails (*Lymnaea stagnalis*); more specifically, the memory to hold their breath and not extend their breathing tubes above the surface when the water in which they reside is low in oxygen. Explaining that one molecular system – the endocannabinoid system – in our brains is thought to be a key player in mediating stress, disrupting our ability to form memories, Lukowiak, Hiroshi Sunada and their colleague Etsuro Ito, from Waseda University, Japan, wondered whether their humble pond snails also used the same system and, if so, how it affected their ability to form memories. But first they had to find the receptor molecules that could mediate endocannabinoid effects in the snails.

Searching a database of the pond snail's genes with the sequence of a cannabinoid receptor that had been identified in the vase

tunicate sea squirt, Takayui Watanabe, from the University of Sapporo, and Dai Hatakeyama identified two candidates, before confirming that both of the genes are expressed to produce the necessary receptors, not only in the snail's nervous system, but also in other tissues. In addition, they found more of the second receptor produced in the nervous system than the first receptor, which was more widely distributed throughout the snail's body. Next, the team investigated whether the receptors could disrupt the snail's ability to form memory.

Injecting the snails with a drug that is known to trigger the endocannabinoid receptor and disrupt memory formation in mammals, Sunada, Sangmin Lee and Jeremy Forest then trained the molluscs to keep their breathing tubes closed when their water was deoxygenated – by poking the breathing tubes with a toothpick whenever the snails tried to stick them out of the water – to find out whether the snails could still form a memory: but they did not. And when Sunada, Lee and Forest tested whether a traumatic experience would also block the snails' memories – by jabbing them on the foot before breath-hold training – the snails were equally unable to learn and form the new memory. However, when the team administered a drug that could block the endocannabinoid receptors from disrupting memory formation 1 h before traumatising the snails, this time the snails were able to learn to keep their breathing tubes closed when the water was deoxygenated and they wanted to pop up for air.

The stressful jabbing experience had the same effect on the snails' memories as the drug that triggered the endocannabinoid receptor, making it likely that the endocannabinoid receptor was activated by stress to disrupt memory formation. Lukowiak says, 'We feel pretty confident that this [the endocannabinoid] system in the snail plays a major role in determining how stress alters memory formation', and he adds, 'We think that it plays an important role in determining the "state" of the animals and thus whether a specific stimulus will be regarded as a stressor'.

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Sunada, H., Watanabe, T., Hatakeyama, D., Lee, S., Forest, J., Sakakibara, M., Ito, E. and Lukowiak, K. (2017). Pharmacological effects of cannabinoids on learning and memory in *Lymnaea*. *J. Exp. Biol.* **220**, 3026-3038.

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High and dry limpets do better when the heat is on



Limpets (*Lottia digitalis*) on a rock. Photo credit: Brittany Davis.

Nothing in nature is constant, from the grand seasonal cycles that sweep the higher latitudes to the daily solar rhythm of tides that leaves exposed coastal residents roasting in the sun before they are plunged again beneath the waves. 'Aerial exposure and the unpredictable nature of the thermal stress during low tides may be critical factors in defining the thermal physiology of intertidal organisms', says Anne Todgham, from the University of California, Davis, USA. However, Todgham points out that few lab-based studies take into account the random thermal fluctuations that shore dwellers experience in their natural environment: lab-based animals are generally exposed to a single heat wave after a period of stable preparation in the lab. Wondering how shoreline residents cope with the natural variation in their thermal regime, Todgham and her colleagues recreated the unpredictable climate that fingered limpets (*Lottia digitalis*) clinging to the Northern California shore experienced during the summer months between April 2011 and March 2013 to find out how daily temperature fluctuations affect their thermal tolerance.

Working with Madeline Drake and Nathan Miller, Todgham recreated the low-tide conditions at Fort Ross with a heating block while washing fresh seawater over the gastropods to simulate high tide twice a day for a fortnight. In addition, the team exposed limpets to: a reliable tidal cycle (where they were always warmed to 24°C); a second scenario where they were warmed repeatedly to 32°C (the highest temperature recorded at the Fort Ross site); a third scenario where the tide washed back and forth, but the limpets experienced no warming during low tide; and a final scenario where the limpets

were continually submerged at 13°C. After completing the 14 d tidal simulations, Drake and Miller measured the heart rates of a selection of the limpets as they were warmed gently (6°C h⁻¹) until the temperature threatened their survival and their heart rates dropped suddenly. The team also collected portions of the limpets' bodies and analysed them for evidence of stress.

Impressively, the limpets that had experienced the Northern California simulated tide were best prepared as the temperature began to rise, with their hearts holding out until the temperature reached ~40.5°C. Meanwhile, the limpets that experienced reliable tidal cycles (where the low tide temperature never varied) held out to ~38°C. In contrast, the hearts of the limpets that had been immersed for the entire fortnight began to fail at ~36°C. Todgham says, 'Repeated aerial exposure alone (the most predictable aspect of the tidal cycle), regardless of the magnitude of temperature increase, had the largest effect on maintaining a high upper temperature tolerance in limpets', adding, 'the heating during low tide is less important'.

However, when Drake analysed how the limpets managed their energy budget and resistance to stress, she and Todgham were surprised to find that the limpets that had experienced the most variation in their conditions were no better prepared than the limpets that had been submerged for the entire period.

So it seems that repeated exposure to the air at low tide is critical for the limpets' high temperature preparations, and Todgham says, 'If aerial exposure is the predominant factor driving thermal tolerance of intertidal limpets, and perhaps intertidal organisms more broadly, this suggests that organisms inhabiting the low intertidal are likely sensitive to warming not only as a result of a thermal history of lower temperatures but also as a result of not being predictably exposed to air during low-tide periods'.

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