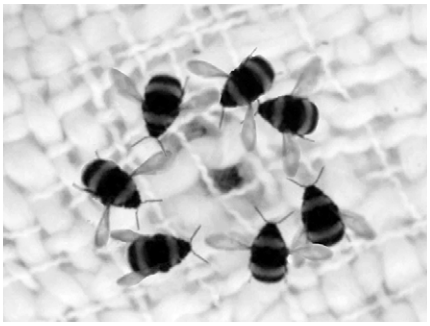
© 2014. Published by The Company of Biologists Ltd | The Journal of Experimental Biology (2014) 217, 2621-2623

INSIDE JEB

How bumblebees detect near and far


Bumblebee every 0.3 s at the start of learning flight. Photo credit: Natalie Hempel de Ibarra.

The first time that a bumblebee leaves the nest and takes to the air, the stakes are high: lose your way and you could be lost forever. The pressure is on to learn as much as possible about the lay of the land before venturing further afield. ‘Bumble bee nests are hidden in the undergrowth’, says Tom Collett from the University of Sussex, UK, adding, ‘The bees have to learn the exact relationship between objects that define the position of the nest and the nest hole’. So instead of embarking on an epic journey and keeping their limbs crossed for a safe return home, the novices set about exploring the vicinity. First, they fly tiny looping circuits that are centred on the nest, gradually broadening the survey until they have learned enough about their surroundings to guide themselves home at the end of their maiden flight.

‘One thing that they need to know is whether objects are near or far’, says Collett. He explains that bees, and insects in general, use the speed that the image of an object travels across the retina of the eye to estimate their distance to an object: images of nearby objects move much faster than images of distant objects. Insects often simplify the task of estimating distance by making sure that the head does not rotate, compensating for any body rotation by moving the head in the opposite direction to the body’s rotation. But bees may use a different strategy to estimate the distance separating two objects, such as the nest entrance and nearby foliage. They could circle around the nest so that objects that are near by move slowly across the eye, while more distant objects move faster. Wondering how bumblebees learn the layout of objects surrounding the nest, Collett and colleagues Olena Riabinina, Natalie Hempel de Ibarra and Andrew Philippides set about filming the insects’ looping learning flights (p. 2633).

The team set up a bumblebee nest box, complete with a queen and several dozen workers, on the roof of a building at the University of Exeter, and then filmed the first tentative flights of new bees as they emerged from the nest. The insects initially strayed no more than a few centimetres from the nest entrance, but they eventually flew out of camera view 20–30 s later. Then the team began the painstaking task of analysing how the learners moved their heads and bodies to find out whether they moved their heads to compensate for body rotations to stabilise the image as they circled around the nest. First, they measured the position of the bee’s body in each frame of the movie as it circled the nest and then they meticulously measured the head movements by hand.

‘We didn’t know what we were going to get’, laughs Collett, but eventually the team was surprised to see that instead of stabilising the image rotation completely by pivoting the head to counteract the body rotation, the learners’ head movements under-compensated for the body rotations, allowing their heads to rotate slightly. Calculating how images move across the retina as the head rotated a little, the team could see images of objects close to the entrance of the nest would move slowly, while remote objects would be moving faster, allowing the bee to distinguish between landmarks that were remote from the nest and those that would guide them home safely.

doi:10.1242/jeb.111088

Riabinina, O., Hempel de Ibarra, N., Philippides, A. and Collett, T. S. (2014). Head movements and the optic flow generated during the learning flights of bumblebees. *J. Exp. Biol.* **217**, 2633-2642.

Kathryn Knight

HP complex is not an essential hibernation switch

Switching off for winter is not easy. Only a select few species dramatically reduce their metabolism and hibernate to endure the long dark months, and how this remarkable tactic came about is not clear. ‘The evolutionary origin [of hibernation] is completely unknown’, says William Wong from Johns Hopkins University School of Medicine, USA. According to Wong, a team lead by Noriaki Kondo from the Kanagawa Academy of Science and Technology, Japan, discovered a protein complex – hibernation protein (HP) complex – in 1992 that was present in the genomes of hibernating squirrel species, such as chipmunks, but absent in non-hibernating squirrel species. ‘This implies the acquisition of unique genes in the course of evolution that enables mammals to hibernate’, says Wong. However, it wasn’t clear whether the presence of this complex was restricted to hibernators. Could the complex turn up in the DNA of non-hibernating mammals too? ‘Given that the genomes of many mammals have been sequenced, we decided to see if HP complex is indeed unique to the hibernators’, says Wong (p. 2667).

However, when Wong and his colleagues searched the genomes of non-hibernating mammals, from the nine-banded armadillo and European rabbit to the bottlenosed dolphin, cow, pig and elephant, they were astonished to find that all of the genes that encode the HP complex were also present in the genomes of these animals. In addition, they found that the *HP* genes occurred in the same locations in the genomes of non-hibernators as they did in the hibernators’ genomes. So, the genes had been conserved during evolution even though none of the species under investigation were hibernators, but how were the genes functioning in the non-hibernators? Maybe there was something different about the way that the genes were expressed to

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.

prevent the non-hibernators from hibernating.

To test this, Wong used cow blood and cerebrospinal fluid samples, provided by his colleague Martin Groschup from the Friedrich-Loeffler-Institute, Germany, that had been collected every spring, summer and autumn over a 4 year period. 'This allowed us to test if the cow HP complexes also oscillate in a seasonal manner analogous to chipmunk HP complexes', explains Wong. Having used antibodies that recognised the HP complex protein to measure the protein's levels in both body fluids, Wong admits that he was impressed to see that the complex peaked in the cow's cerebrospinal fluid in February, just like in the hibernating chipmunks. The team also showed that the complex was produced by the cow's liver, was composed of three HP proteins and that each protein was modified with sugar molecules, just like in the HP complex in chipmunk blood. And when the team tested whether the HP complex could produce hibernation effects when injected into mice, they found that the rodents ate less, although they did not drop into hibernation.

'The *HP* genes are not unique to hibernators', says Wong, who adds, 'They are conserved in non-hibernating mammals and the HP complexes likely regulate physiological functions distinct from hibernation'. Wong cautiously suggests that the complex may regulate food intake in non-hibernators, but emphasises that this result needs to be retested in an animal that possesses *HP* genes. And, having shown that the HP complex is not the essential switch that throws animals into hibernation, Wong says, 'Until hibernation-specific genes are found, it is more likely that differential gene expression and/or re-wiring of existing endocrine circuits enable hibernators to hibernate'.

doi:10.1242/jeb.111096

Seldin, M. M., Byerly, M. S., Petersen, P. S., Swanson, R., Balkema-Buschmann, A., Groschup, M. H. and Wong, G. W. (2014). Seasonal oscillation of liver-derived hibernation protein complex in the central nervous system of non-hibernating mammals. *J. Exp. Biol.* **217**, 2667-2679.

Kathryn Knight

Rhodnius respiration depends on multiple factors

Fed and unfed *Rhodnius prolixus*. Photo credit: Timothy Bradley and Catherine Loudon.

Sucking great lungfuls of air into the body is the main response of most mammals to physical exertion. As their metabolic rates rise, they inhale more deeply to maintain power. However, for insects, breathing is more complex. Equipped with a network of fine ventilation tubes that permeate every tissue in the body, insects have to tightly regulate the passage of oxygen into the body as their metabolic demands increase, by altering their breathing patterns. Timothy Bradley from the University of California, Irvine, USA, notes that respiratory patterns were thought to be principally determined by metabolic rate. But bugs don't just ramp up their metabolism in response to exercise; their metabolic rates increase as temperatures rise and during reproduction and digestion, so it wasn't clear whether other activities might result in different respiratory patterns. 'There was some evidence that the respiratory control mechanism might be temperature sensitive', says graduate student Erica Heinrich. She and Bradley began investigating the effects of feeding and temperature on the blood-sucking insect *Rhodnius prolixus* (p. 2752).

'*Rhodnius prolixus* provides certain valuable features as a model insect for studying respiratory control', says Heinrich, who explains that the insect's metabolic rate rockets after dining on blood. This allowed her to measure the metabolic rates and respiratory patterns of the insects as they exhaled CO_2 after a satisfying rabbit blood meal, in addition to monitoring the effects of temperature on the unfed insects as she varied the temperature between 18 and 38°C.

Sure enough, as the temperature rose, the insects' metabolic rate increased 3.5 times above their resting metabolic rate and, as their metabolic demands increased, they initially opened and closed their spiracle valves (breathed discontinuously) until the metabolic rate became high enough and the spiracles remained open continuously.

Next, the duo measured the metabolic rates of the well-fed insects and they were amazed to see the bugs' metabolic rates hurtle to almost 14 times their resting metabolic rates. However, when they analysed the bugs' CO_2 exhalation pattern, they found, surprisingly, that instead of holding the spiracles open and exhaling CO_2 continuously, they continued closing the spiracles intermittently, even though the insects' metabolic rate was four times higher than that at the hottest temperature.

The duo also monitored the amount of CO_2 released by the insects during each exhalation burst and found that they exhaled less CO_2 per breath as the temperature rose. This was in contrast to the digesting insects, which exhaled more CO_2 each time they opened their spiracles as their metabolic rates rose.

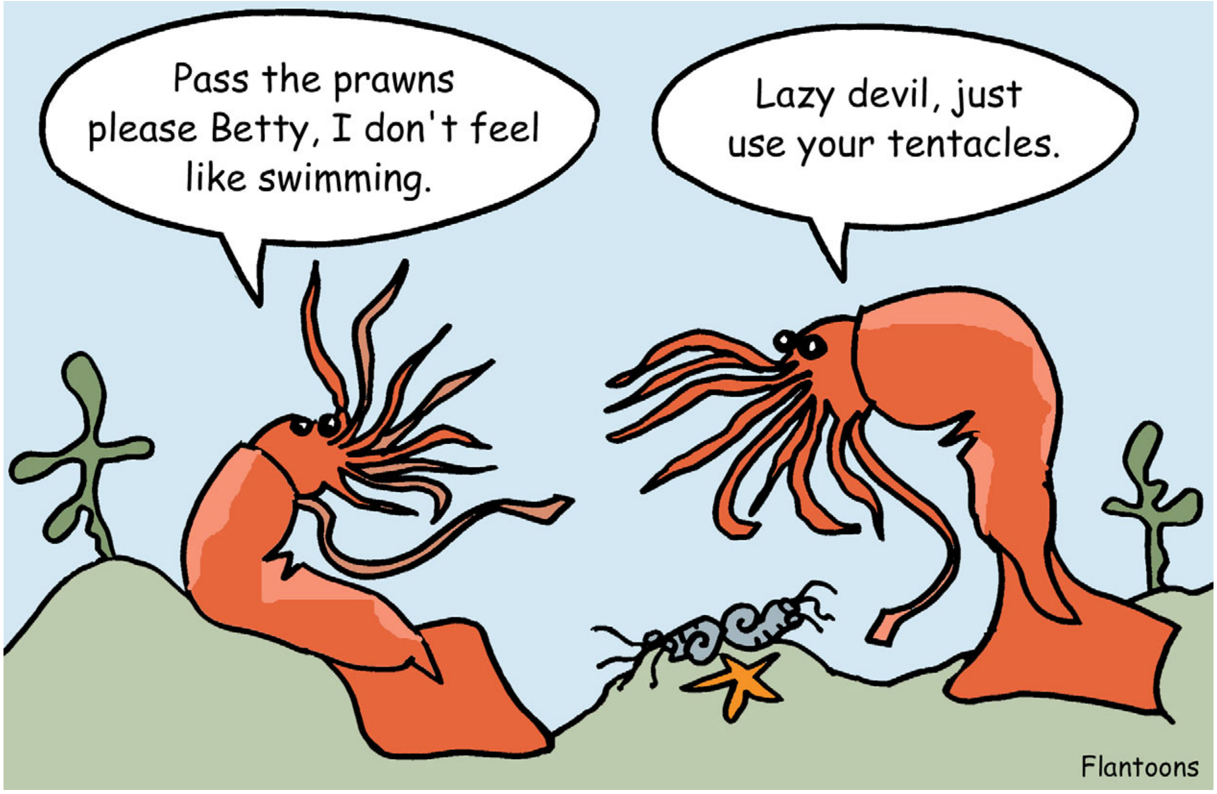
Having shown that the insects' respiratory pattern was different under the two situations, Bradley says, 'It is overly simplistic to attribute respiratory pattern to metabolic rate alone', and the duo suspects that the insects' respiratory system is temperature sensitive. Explaining that if the respiration system was not sensitive to temperature, insects should always exhale the same amount of CO_2 each time that open their spiracles, Heinrich says, 'However, we found that as the temperature increases, the volume of CO_2 released in a burst decreases'. She and Bradley suspect that this temperature sensitivity may result from changes in the pH of water in the insect's body. 'Increased temperature decreases the pH of water', explains Heinrich. She adds, 'If the CO_2 threshold that triggers spiracle opening is sensed via pH, then exposure to high temperatures will trigger premature spiracle opening… and less CO_2 would need to accumulate via metabolism to reach the pH threshold. This will result in reduced volumes of CO_2 release during spiracle opening'.

doi:10.1242/jeb.111104

Heinrich, E. and Bradley, T. (2014).Temperature-dependent variation in gas exchange patterns and spiracular control in *Rhodnius prolixus*. *J. Exp. Biol.* **217**, 2752-2760.

Kathryn Knight

Largest squid have lowest anaerobic metabolic capacities



Hunting beneath the waves comes with its own unique set of challenges. As swimming predators grow, the drag holding them back increases when they speed up, and the chances of being detected by their victim also increase, so stalkers have to mount an even greater burst of speed to intercept prey. This requires that predatory fish increase their metabolic capacity as they grow. But how does the metabolism of squid that reside in the same environment alter as they increase in size: do their metabolic capacities increase as they grow? Intrigued, Lloyd Trueblood from La Sierra University, USA, and Brad Seibel from the University of Rhode Island, USA, went trawling and fishing for two species of squid – *Dosidicus gigas* ranging from 0.16 to 17,200 g and the more diminutive *Doryteuthis pealeii* (7–135 g) – to find out how their anaerobic capacity varied with size (p. 2710). The duo was amazed to discover that instead of increasing their anaerobic metabolic capacities, the largest animals from both species had the lowest capacities for their size.

Trueblood and Siebel suggest that the largest squid have reduced anaerobic metabolic capacities because they may not need to swim as fast as predatory fish when hunting; instead, they can reach for prey with their tentacles. They suspect that approaching more stealthily and striking from farther away also accounts for the squid's unusual metabolic characteristics. In addition, the duo explains that *D. gigas* largely dine on the same sized prey throughout their lives – unlike fish, which pursue larger and faster prey as they grow – so that their burst speed and power might decline because their prey don't speed up as the squid grow: 'They can get away with being lazy', says Trueblood.

They also explain that older *D. gigas* are protected from predation as fish tend to dine on prey that are 10–20% their own size, leaving larger *D. gigas* alone, and add that *D. gigas* migrate for part of each day down into the oxygen minimum zone, where they are protected from sharks and other top aquatic predators that cannot survive in the hypoxic conditions. The duo suggests that these factors could all have contributed to the reduction in the squids' anaerobic capacity as they grow.

doi:10.1242/jeb.111112

Trueblood, L. A. and Seibel, B. A. (2014). Slow swimming, fast strikes: effects of feeding behavior on scaling of anaerobic metabolism in epipelagic squid. *J. Exp. Biol.* **217**, 2710-2716.

Kathryn Knight
kathryn@biologists.com