Keeping track of the literature isn't easy, so Outside JEB is a monthly feature that reports the most exciting developments in experimental biology. Short articles that have been selected and written by a team of active research scientists highlight the papers that JEB readers can't afford to miss.

Outside JEB

LONG LIFESPAN

THE DEADLY SCENT OF FOOD!

It is a well-known phenomenon, from *C. elegans* to lab rat to human, that severe dietary restriction greatly extends life span. Of course, the problem is that even if severe dietary restriction *didn't* extend life span, it would certainly feel like it, as eating becomes a joyless experience. This is why 'big pharma' is trying to identify drugs that interfere in the pathway, so that we can eat as much as we like and *still* live longer. How does the body sense the reduction in nutrient quality, and what is changed? Downstream elements of the pathway seem to include insulin signalling, but which sense is involved is less clear.

Recent work in *C. elegans* suggested a possible role for taste in longevity, and so Scott Pletcher and colleagues decided to apply *Drosophila* genetics to the question, because behavioural analyses of olfaction and feeding are also relatively advanced in *D. melanogaster*. As this tiny fly naturally feeds on rotting fruit, it finds the volatile odours generated by yeast to be particularly appealing. However, a little of what you fancy can actively do you harm, as we shall see. The authors exposed flies on a severely restricted diet to these strong-smelling, volatile odorants from live yeast, or live yeast itself. Both treatments significantly reduced life span, showing that odorants alone are capable of restricting life span. Are these odorants simply toxic? Apparently not, because flies fed on normal diet are unaffected by the same odorants. So it seems as if at least a part of the life extension caused by dietary restriction is attributable to odorant-mediated perception of restricted nutrient availability.

If this were the case, then genetic blockade of olfaction might be expected to extend life span. In *Drosophila*, olfaction is mediated by a family of 62 odorant receptors. One of these, *Or83b*, is broadly expressed and is required for correct targeting of other receptors. Mutants are almost anosmic, meaning that they can't smell. Fully-fed female mutants of *Or83b* lived over 50% longer than wild-type flies, and males also lived longer, although the difference was less spectacular. This increased life span was 'rescued' back to normal when a normal copy of *Or83b* was expressed in *Or83b* mutants, showing that the longevity effect was a direct effect of *Or83b* mutation. In simple terms, these results would suggest that the smell of rich food restricts life span.

What happens when *Or83b* mutants are exposed to dietary restriction? In fact, the enhanced longevity effect is maintained even under conditions of severe restriction. This shows that olfaction is not a necessary step for the effect of dietary restriction, although there is obviously some interaction between their sense of smell and the effect of eating less. Presumably the fly has some other means of detecting nutritional quality of food, such as sensing the level of nutrients in the haemolymph, so the olfactory pathway is semi-redundant.

Nonetheless, the depressing message from *C. elegans* and *Drosophila* is clear: if you want to live longer, eat much less and don't enjoy it.

10.1242/jeb.000836

Sergiy Libert, S., Zwiener, J., Chu, X., VanVoorhies, W., Roman, G. and Pletcher, S. D. (2007). Regulation of *Drosophila* life span by olfaction and food-derived odors. *Science* **315**, 1133-1137.

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FLIGHT STABILITY

HOVERING IN THE DARK

In spite of the huge selective advantages that it can give an animal, powered flight has evolved only four times in the history of life on Earth: in insects, pterosaurs, birds and bats. One of the things that makes powered flight complex in animals is that it has to be accomplished via flapping, which generates inherently unsteady aerodynamic forces as the wings move back and forth. These challenges are greatest for hovering animals, which require an intricate feedback system to remain in one place. In dragonflies, flight stability is probably accomplished using visual feedback from the eyes. In other insect groups, one pair of wings has been modified into a pair of club-shaped structures called halteres. These structures allow two-winged insects such as flies and mosquitoes to detect rotational movements of their bodies, which is critical for the maintenance of flight stability in the face of external perturbations like gusts of wind.

In a recent article in *Science*, Sanjay Sane and colleagues ask the question of how moths achieve flight stability in the absence of both visual cues and halteres, since moths fly in low-light conditions and have two pairs of functional wings. In this paper, they test the hypothesis that hawkmoths (*Manduca sexta*), which are excellent hoverers, use their antennae in the same way that flies use their halteres. This hypothesis predicts that moth antennae, like halteres, should vibrate at a rate that is comparable to the wing beat frequency. This is exactly what they found when they used high-speed video to analyze the motion of moth antennae during hovering. By recording from neurons in the base of the antennae, they also demonstrated that antennae are capable of detecting the kinds of mechanical perturbations that they would experience as a result of bodily rotations during hovering flight.

While these experiments showed that the antennae could act as rotation sensors, they did not demonstrate that they actually do. To directly measure the contribution of the antennae to flight stability, the researchers amputated antennae from a group of moths and measured their flight performance. Moths lacking antennae were far more likely to crash to the ground or into the walls of the test chamber, which was compelling evidence that the antennae contribute to flight stability. Antennae are endowed with a rich diversity of sensory neurons that detect not only mechanical input but also chemicals, humidity and temperature. To test whether any of these sensory systems is involved in flight stability, the investigators re-attached antennae with cyanoacrylate glue and found that this rescued flight performance.

These results lead to two important conclusions. The first is that the odor, humidity and temperature sensors are not involved in flight stability, since the axons from these structures were still severed in moths with re-attached antennae, and these moths could still fly. The second is that the mechanical sensors used for flight stability must be located at the base of each antenna, below the point where they were cut. Structures called Johnston's organs are located in this area and are known to detect antennal movements associated with sound perception, so it is likely that the Johnston's organs are also used in flight stability in moths. These findings are interesting because they suggest that although the evolution of powered flight has been a rare event, mechanisms of achieving flight stability have evolved several times in insects using a variety of sensory structures including the eyes, wings and antennae.

10.1242/jeb.000844

Sane, S. P., Dieudonné, A., Willis, M. A. and Daniel, T. L. (2007). Antennal mechanosensors mediate flight control in moths. *Science* **315**, 863-866.

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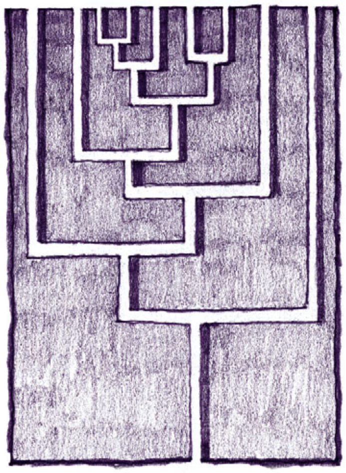
TOOTH WEAR



LONG IN THE TOOTH

When a herbivore wears away its teeth to nothing by years of chewing on abrasive foliage, its outlook is not bright. Many specialist herbivore species have independently evolved long teeth, often as part of an arms race with plants armouring themselves with silica. It is then tempting to suppose that longer teeth should lead to longer life spans; however, recent work by Vebjørn Veiberg and colleagues provides strong evidence that the reverse can be the case, with longer life-span requirements driving the evolution of longer teeth.

The research team studied two populations of roe deer living wild in forests in France. Both live in habitats dominated by oak and beech, but the forests differ in climate and productivity. Deer from the population living in the less productive environment tend to have a longer life expectancy, probably due to caloric restriction. On the other hand, those deer from the more productive forests tend to follow the 'live fast, die young' strategy. They are probably more capable of producing more offspring at a younger age but also suffer a higher mortality rate.

The team wanted to test two hypotheses: the 'tooth-wear' hypothesis, which relates variation in tooth height to diet quality, and the 'life-history' hypothesis, which relates initial molar height to life expectancy. They measured molar heights – top of tooth to bottom of enamel – in 93 animals found dead within the study areas and correlated this with age. Veiberg's team show no difference in the *rate* of tooth wear between the two populations, but deer from the poor quality environment, with long life expectancies, started adulthood with longer molars despite being generally smaller animals. Thus, differences in tooth height are related to adaptations for different life expectancies rather than diet abrasiveness or rate of chewing. Deer from poor habitats have to survive long enough to reproduce effectively despite their low-calorie diet; increased initial tooth height evolves in response to this longer life-span strategy, supporting the life-history hypothesis.

While it makes some sense that individuals from a population with greater life expectancies have to be better built, this study is remarkable in that it isolates life expectancy from other factors exceptionally well, which is challenging to show in wild populations of relatively large mammals. Previous work by Juan Carranza and colleagues, published in *Nature* in 2004 (vol. 432, p. 215), related tooth wear to life expectancy and suggested a relationship between life expectancy and initial tooth height in red deer. The males truly do ‘live fast, die young’: not only do they have smaller teeth than females despite their considerably larger size but they also expend huge amounts of energy competing with other males to mate with females. Hence, they eat much more and wear their teeth down faster, making it difficult to attribute tooth height to life expectancy. Therefore, Carranza and colleagues couldn’t reject the tooth-wear hypothesis. Veiberg’s work on roe deer, by contrast, compares populations with very similar tooth wear rates, so that the tooth-wear hypothesis can be cautiously rejected.

So, what we have here is in nice agreement with ageing evolutionary theory, but it is also vaguely disturbing. That such a simple metric as initial adult tooth height might give away an organism’s ‘design life-span’, or effective ‘warranty period’, has interesting implications. What if insurance companies could determine a similar metric for humans? Instead of looking at indicators of risk for some specific failure (such as heart disease), they could look at an apparently innocent trait and calculate our allotted life spans.

10.1242/jeb.000877

Veiberg, V., Mysterud, A., Gaillard, J.-M., Delorme, D., Laere, G. V. and Klein, F. (2007). Bigger teeth for longer life? Longevity and molar height in two roe deer populations. *Biol. Lett.* **3**, 268-270.

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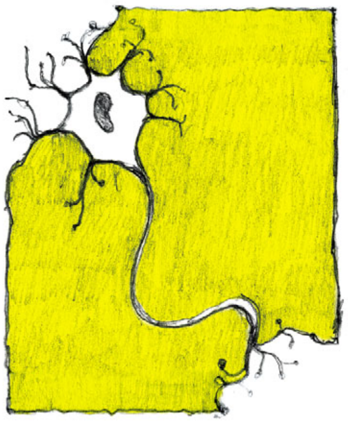
TURNING CONTROL



COCKROACHES GOING ROUND THE BEND

For animals making their way through complex environments, walking is not only a question of moving their legs in rhythmic patterns, but they also may need to avoid, obstacles or to reach across gaps. Many insects are exceptionally good at maneuvering through complicated environments such as the forest canopy, where maintaining coordination is essential since the penalties for poor performance can be high: failing to find mates, food or, worse still, becoming someone else’s food! Although much is known about how insects control the movements of a single limb, how they maintain coordination between limbs whilst negotiating obstacles or gaps is less clear. To do this, insects must take sensory information obtained by rhythmic movements of the antennae or forelimbs as well as by the visual system and use this information to plan leg movements during walking, which is difficult to investigate experimentally.

Angela Ridgel and colleagues at Case Western Reserve University in Cleveland used brain lesions in cockroaches, *Blaberus discoidalis*, to investigate the role of one enigmatic brain region that has been implicated in controlling limb coordination during obstacle avoidance and turning: the central body complex (CBC). Following 24 h recovery after surgery, the team assessed the cockroaches’ behavior in two setups designed to make them turn. Using a U-shaped arena, they could assess more general turning and obstacle-avoidance behavior, while using a tethered setup in which a wall was moved to simulate an obstacle allowed them to examine the detailed movements of the legs.

Lesions made with a razor blade, which severed links between the left and right sides of the CBC or connections to and from one side of the brain and the CBC, had severe effects on the turning behavior of the cockroaches in the U-shaped arena. Over half the turns made by these animals were abnormal, including turning in circles, turning towards stimulation of one antenna but away from stimulation of the other or simply running into the wall. The team also made foil lesions at several regions within or closely associated with the CBC and likely to be involved in walking. In many cases, cockroaches with these lesions failed to turn and ran straight into the wall. This showed that lesions made with a razor blade mainly disrupt left–right coordination – the cockroaches still turn but do so incorrectly – whereas foil lesions prevent the cockroaches turning at all. More detailed analysis in tethered cockroaches showed that many of the lesions made with foil or razor blades prevented cockroaches from producing the appropriate leg movements to turn and avoid an obstacle.

As the authors point out, this study represents only the beginning of studies aimed at finding out how the CBC and its closely associated brain regions contribute to limb coordination during complex behaviors. Their results already emphasize the importance of communication between the two sides of the CBC for turning in the right direction and avoiding obstacles and of communication between the CBC and other parts of the brain in allowing the cockroaches to turn at all. The next step for Ridgel and colleagues may be to delve further into the CBC’s circuits in the brain, where they may find answers to many of the questions the present study raises such as how the CBC integrates inputs from the left and right antennae to adjust the motor programs that control walking and initiate turning. These experiments are likely to prove challenging but have great potential for providing new insights into insect motor control.

10.1242/jeb.000869

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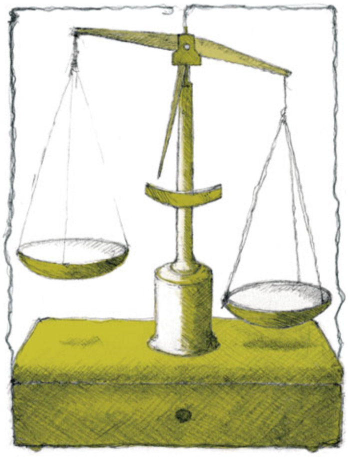
ANOXIA SURVIVAL



SENSATIONAL SHELLS

In contrast to the vast majority of vertebrates, which usually die within minutes when deprived of oxygen, freshwater turtles are extremely resistant to oxygen deprivation, or anoxia. However, not all freshwater turtles are equally capable of surviving anoxia. For instance, the champion survivor, the Western painted turtle, can recover from up to 100 days of anoxia at winter temperatures (3–5°C), whereas the red-eared slider turtle succumbs after around 45 days of anoxia at the same temperature. Why such differences in anoxia survival time exist between turtle species remains a mystery, although Donald Jackson's team at Brown University has been working feverishly on elucidating what factors may account for the differences in anoxia survival times.

In their recent paper, the team surmises that, since all freshwater turtles are resistant to anoxia *per se*, differences in anoxia survival time among turtle species must be due to how effectively a turtle can extend the time before the changes in physiology that accompany anoxia become irreversible and lethal. For example, turtles' fuel reserves, such as liver glycogen, become depleted and the harmful acidic end products of anaerobic metabolism, lactate and H^+, accumulate. Therefore, the greater anoxia tolerance of some turtles compared with others could be due to more anoxia-tolerant species exhibiting traits that prolong the time until glycogen reserves are exhausted and/or a critically acidic pH is reached.

Innovatively, a turtle's shell is not just protective armour but also reduces the harmful effects of H^+ accumulation during anoxia exposure. Specifically, shells release calcium, magnesium and sodium carbonates into the extracellular fluid in exchange for lactate to supplement extracellular buffering of H^+. Thus, Jackson's team hypothesized that some of the difference in anoxia tolerance between turtle species could be due to differences in buffering characteristics of their shells.

To test this, the team made a number of measurements to test how well the shells of different turtle species could contribute to acid buffering. First, they compared shell mineral composition and total shell CO_2 concentration, which is related to the amount of carbonate in the shell, to tell them how many ions were potentially available for buffering. They found that shells from the more anoxia-tolerant painted and snapping turtles had more mineralised shells and more shell CO_2 than shells from the less anoxia-tolerant map, musk and red-eared slider turtles, suggesting that anoxia-tolerant turtles had more ions available for buffering.

Next, the team tested if the amount of buffer chemicals released from the shell differed among species. They measured the amount of acid they needed to add to powdered shell samples before the solution pH could no longer be maintained constant at pH 7 and found that more acid was needed for shell samples from more anoxia-tolerant species, indicating that these shells could release more buffer. Finally, the team tested whether there were differences in the shells' ability to accumulate lactate. They incubated pieces of shell in a standard lactate solution for a set time period, finding that shells from anoxia-tolerant turtles accumulated more lactate.

Because shells of more anoxia-tolerant turtle species contain more minerals and CO_2, release more buffer chemicals and accumulate more lactate than those of less anoxia-tolerant turtles, the team concludes that differences in shell composition and buffering properties likely account for some of the reported differences in anoxia tolerance among freshwater turtles. The superior shell-buffering mechanisms of the more anoxia-tolerant turtles probably slow the development of highly acidic conditions during anoxia, prolonging survival time.

10.1242/jeb.000851

Jackson, D. C., Taylor, S. E., Asare, V. S., Villarnovo, D., Gall, J. M. and Resse, S. A. (2007). Comparative shell buffering properties correlate with anoxia tolerance in freshwater turtles. *Am. J. Physiol.* **292**, R1008-R1015.

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