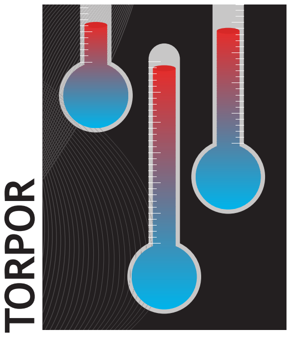
OUTSIDE JEB

Hot bats change torpor game rules



In just the same way that turning off the furnace in your house in the middle of a cold winter night causes the property to cool, when hibernating mammals stop generating heat during winter to conserve energy, their temperature also drops to match that of their environment. But not all animals turn down the inner thermostat for an entire season. A shorter version of hibernation is torpor, where animals stop keeping their body warm for a few hours or days when it is cold or when food is scarce. However, most of our understanding of how animals use torpor has been gleaned from research done in colder regions, where there is little spare heat and an animal's body temperature drops as soon as they turn down their inner furnace. But what happens when animals resort to this form of energy conservation in warmer climes? Stephanie Reher and Kathrin Dausmann, from the University of Hamburg, Germany, recently studied a species of bat (*Macronycteris commersoni*) in Madagascar and found that when these bats slow down their rate of energy usage, their body temperature actually increases.

Reher and Dausmann found this new flavour of torpor by measuring both the bats' skin temperature and metabolism when the animals were resting during the day. Unexpectedly, their skin temperature increased when the bats reduced their metabolism. In other words, the bats had been actively using energy to stay cooler than the surrounding air temperatures, and as soon they stopped putting effort in, their bodies heated up. In fact, the air temperature in the dry forests of Madagascar is often hotter than the bat's body temperature, sometimes as high as 41°C, and the bats' body temperatures even reached 42.9°C on one occasion. Reher and Dausmann think that this 'hot torpor' saves the bats energy and allows them to better tolerate hotter temperatures.

In addition to this unusual form of torpor, the bats did a second unique thing on cooler days, around 34°C; some bats entered many micro-spells of torpor throughout their daytime resting period, almost like cat-napping when you are supposed to be listening to a presentation. During these micro-spells of torpor, the bats used less energy, essentially pausing their metabolism for only a few minutes at a time (3–53 min). Because these micro-spells were so short, the body temperature did not have time to reach that of the local air; it only changed a little, shifting toward the air temperature. The researchers think this penny-pinching attitude saves the bats energy over time, but allows them to remain alert to any threats while they are roosting, compared with traditional lengthier periods of torpor when bats are lethargic or even unconscious – imagine being woken from a very deep sleep.

In addition, this discovery of a novel mode of torpor reveals the importance of studying species in the tropics, where high temperatures can cause major physiological differences relative to species that reside in cooler climes. This finding, that some species do not follow the traditional rules of torpor, may also suggest that the rules were not right to begin with. The fact that body temperatures can increase when the metabolism is reduced may force us to reconsider our previous understanding of torpor and rewrite what, until now, was an accepted rule of thermal physiology.

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Reher, S. and Dausmann, K. H. (2020). Tropical bats counter heat by combining torpor with adaptive hyperthermia. *Proc. R. Soc. B.* **288**, 20202059. doi:10.1098/rspb.2020.2059

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Flight among the Lilliputians

If you think that fruit flies are tiny, please reconsider. Spectacular radiations of miniaturized beetles, flies and wasps dominate the insect fauna and are characterized by body lengths well below that of the biologist's friend *Drosophila melanogaster* (~3 mm). Most remarkably, the smallest flying insect described to date has a length of just 0.16 mm. But just how does flapping flight work at such small scales? The viscous effects of air on a wing increase at smaller sizes and a 1 mm insect moves through the air as would a bumblebee move through mineral oil. Lift forces generated by swirling air around the wing are accordingly impeded given the relative stickiness of the fluid, and alternative mechanisms of force production must be at play. How then do very small insects create the aerodynamic forces necessary to offset body weight and also to generate horizontal thrust to intentionally move about?

Xin Cheng and Mao Sun, from Beihang University, China, took on this question by describing the detailed kinematics of forward flight in the tiny wasp *Encarsia formosa*, which has a wing length of only 0.5 mm. Writing in 1973 in JEB, Torkel Weis-Fogh first described the novel clap-fling aerodynamic mechanism utilized by this species; Cheng and Sun have now built on their earlier computational work on hovering in *E. formosa* to analyze its forward flight kinematics and associated forces, albeit at the humble airspeed of ~0.3 m s^{-1}. They identified a distinctive pattern of wing motions very different from

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that of larger insects (including the fruit fly). First, the wings accelerate backward at very high incidence angles relative to oncoming flow ('impulsive paddling'), and then close up on the right and left sides while at the same time moving upward. Then in the downstroke, the wings quickly rotate about their posterior edges (which Weis-Fogh referred to as the 'fling') and sweep downward and forward, again moving at high incidence angles to the surrounding air.

But what do these odd features of wing flapping yield in terms of force production? Cheng and Sun used computer simulations to reveal two means by which the effects of small size and fluid stickiness were largely overcome. Firstly, the fast backward acceleration of the wings at high incidence to the air produces a large thrust but also some vertical force supporting the body weight. Secondly, the 'flinging' motion produces a low-pressure region of rapidly swirling air which persists through the downstroke, creating the majority of the vertical force required to offset gravity. Thus, the insects employ distinctive wing motions to offset the otherwise formidable effects of increased viscosity. The associated forces last just milliseconds but, at a wingbeat frequency of ~350 Hz, are clearly sufficient for routine flight. They may be even more effective during maneuvers, a topic destined for future investigation.

Miniaturization is one of the dominant themes in insect evolution and largely underpins the high extant diversity of small insects (and also of their parasitoids). This computational study of aerodynamics in one such lilliputian species suggests that additional fluid dynamic novelties await discovery and analysis by flight biomechanists. Fringed wings with much reduced supporting veins are also well known among the miniaturized fliers, but have largely unknown aerodynamic consequences. Often at the mercy of gentle breezes, these insects live in physical, behavioral and ecological domains supremely distant from our own personal experiences. This aerodynamic study should help us to close that conceptual distance.

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Mapping odours to fight malaria

Mosquitos have pestered humans since time immemorial, ruining many a summer evening. If you live in the temperate regions of the world, a scented candle, some insect repellent and lots of clapping may be all you need to keep the critters at bay. But make no mistake, in the tropics, mosquitoes kill more people every year than any other animal, by transmitting a variety of deadly diseases, among them malaria. Mosquitoes have an exquisite sense of smell, thanks to a special neuron on their face that works much like our taste buds, and they find their hosts by following the plumes of carbon dioxide (CO_2) that humans exhale. However, the same neurons in the harmless fruit fly are also affected by other odours, and sometimes they lose their ability to detect CO_2. If the same was true for mosquitoes, thought Stephany Chen and her colleagues at the University of California, Riverside, USA, perhaps there is a certain smell that could make us invisible to mosquitoes and rid humanity of them and their diseases once and for all.

In a first experiment, the team placed a single mosquito in a chamber and recorded the activity of its smell neuron, which increased after a puff of CO_2. Then the insect was given a scent to smell, before measuring the response of the neuron to CO_2 once again. In this way, the team screened a large number of odours and found many that would either decrease the response of the neuron to CO_2, increase it, or cause a sustained activation, during which time the neuron is blind to a new puff of CO_2. The latter category of scents, dubbed 'prolonged activators', represents a promising avenue for the development of new insect repellents. Among them, butyric acid proved to be especially effective: only a 3 s whiff of the stuff would keep the mosquito's smell neuron firing continuously for over 5 min, preventing the nose-blind critters from homing in on us. Butyric acid is already part of the odour-cocktail of human sweat, and because we all emanate different amounts of it, this may explain, in part, why some of us are mosquito candy, while others escape unscathed.

There is a whole world of odours out there, some of which may have the same mosquito-repellent properties as the prolonged activator, butyric acid; but how to find them? As is often the case today, the answer is artificial intelligence. Armed with a long list of odours that were found to affect the mosquitoes' ability to smell CO_2, the team fed the data into a computer that worked out which traits of the molecules' three-dimensional structures were common to the prolonged activators and which set them apart from all other effective odours. Then, these findings were used to train a machine-learning algorithm that, after some time, became really good at recognizing promising new candidate molecules; the software can now map the world of odours and predict which can confuse the mosquitoes' sense of smell. It seems that the team is on the scent of a truly awesome repellent.

Among the many odours in the world, there may be a handful that cause the smell neurons of mosquitoes to fire out-of-control, and thus could cloak mankind from the insects and their deadly bite. Butyric acid, for instance, works wonders to make mosquitoes nose-blind, but the odour is not a particularly lovely one: it smells like rancid butter. Before you find yourself wearing it to your next garden party, maybe wait and see whether artificial intelligence will yield a less pungent solution.

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Chen, S. T., Kowalewski, J. and Ray, A. (2021). Prolonged activation of carbon dioxide-sensitive neurons in mosquitoes. *Interface Focus* **11**, 20200043. doi:10.1098/rsfs.2020.0043

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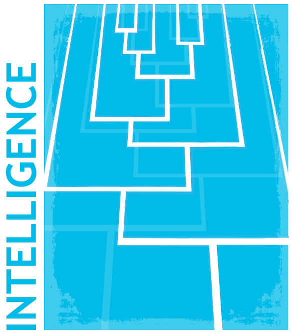
House mice in human habitats are street-smart



I remember the first time I took the New York City subway. The station was vibrant with a mix of passengers waiting for their train and a few subway rodents scurrying away with discarded food. Does living in an urban environment make such animals better at solving challenging tasks to find food? Researchers Lara Vrbanec, Vanja Matijević and Anja Guenther from the Max Planck Institute for Evolutionary Biology, Germany, studied the problem-solving abilities of three subspecies of house mice that have lived in human-altered environments for different evolutionary periods.

The house mouse, *Mus musculus*, has been living with humans for around 13,000 years. During this time, it has evolved into three subspecies: *M. m. domesticus* (western house mouse) appeared 11,000–13,000 years ago, *M. m. musculus* evolved 8000 years ago, and then *M. m. castaneus* turned up approximately 3000 years ago. Although we know the ecological impacts of humans on house mice, our influence on the intelligence of mice was unknown. Animals that can adapt their behaviour to novel circumstances are likely to be more successful in changing environments. So, the scientists wondered whether the mice subspecies that have lived in close proximity with humans the longest are better at solving problems, and can invent novel behaviours or reuse pre-existing ones, than the newer arrivals.

To test their idea, the researchers tested laboratory-based colonies of all three subspecies, which had lived in captivity for several generations after sharing their homes with humans for thousands of years. The researchers set the mice seven tests where they had to access a mealworm reward in different contexts by performing specific manoeuvres, such as pushing, pulling or digging. The scientists found that the captive western house mice were most likely to solve the food extraction tasks, followed by *M. m. musculus* and *M. m. castaneus*. As there were no recent differences in the animals' lifestyles, their different aptitudes could only be accounted for by the amount of time they had spent sharing their homes with humans in the past. The better problem-solving ability of western house mice is probably a result of sharing their living space with humans for the longest time.

However, the team needed convincing that the impressive abilities of the western house mouse are due to their proximity to people and not to some other factor, such as their general inquisitiveness. To test whether any other aspects of their lives drove the animals' increased intelligence, the researchers connected a cage containing unfamiliar objects to the cages of individual mice and measured how long it took each one to enter the unfamiliar environment. They thought that if the subspecies differed in their motivation to enter the unknown environment, their curiosity could be a possible explanation for why the mice vary in their problem-solving abilities. But they found that the three subspecies did not differ in their inquisitiveness or motivation to explore novel objects, confirming that the western house mouse is better at solving challenging tasks likely owing to its longer cohabitation with humans.

Whether you are a New Yorker or a mouse that evolved in an urban or human habitat, problem-solving is key for survival. The human environment may act as a filter in which individuals with certain behaviour types are more likely to thrive. So, the subspecies of mice that has co-existed longest with humans have adapted well to their new, man-made environment through better problem-solving abilities than subspecies that have lived in our shadow for shorter times. So be warned, it may not be long before some mice learn how to extract cheese successfully from your mousetrap without getting squished.

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Vrbanec, L., Matijević, V. and Guenther, A. (2021). Enhanced problem-solving ability as an adaptation to urban environments in house mice. *Proc. R. Soc. B.* **288**, 20202504. doi:10.1098/rspb.2020.2504

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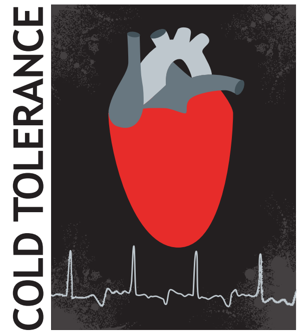
A steady tick for a cool alligator heart



Alligators are cold-blooded reptiles that retain their body heat by seeking out warm environments. But body temperature changes can be a dangerous prospect for the heart. A healthy heart maintains a steady beat by propagating electrical signals throughout the tissue, but low temperatures interfere in this process by changing the electrical capacity of the muscle, possibly causing lethal irregular heartbeats. To learn what keeps alligator hearts safely ticking at low temperatures, Conner Herndon and his colleagues at Georgia Tech in Atlanta, Georgia, USA, set out to investigate why alligator hearts appear to handle temperature fluctuations much better than those of other species, such as mammals, that must maintain a constant body temperature.

Herndon and the team collected heart tissue from several American alligators in order to test how well electrical signals pass through the muscle. The team then trained high-frequency cameras on the hearts and stimulated them with electrodes at a range of heartbeat frequencies to calculate the speed of the electrical signal and how far the signal from one heartbeat travelled across the tissue at temperatures ranging from 23°C to 38°C. In addition, the team repeated the experiments on the hearts of rabbits – an animal that must maintain a constant body temperature to survive – to find out how well both hearts coped at low and high temperatures.

At low body temperatures (23°C), the alligator hearts maintained function much better than the rabbit's: the electrical propagation speed only dropped by 20% in the alligator heart while the rabbit experienced a 67% drop. The distance that the signal travelled in the rabbit's heart shortened to an alarming distance reaching only 3 cm – equivalent to the length of its heart. Shorter stimulus lengths put the animal at risk of heart failure because the muscle tissue farthest away is at risk of being stimulated by stray electrical signals emanating from other sites in the heart, causing an irregular heartbeat. In contrast, alligators maintained a longer propagation distance of 6 cm – double the 3 cm length of their hearts – ensuring full stimulation of the entire muscle tissue and well-coordinated heartbeats at low body temperatures.

The team also found that the alligators' low heart rate helped protect it against the effects of cooler body temperatures. The alligators' maximal heart rate only dropped from 70 beats min^{-1} at 38°C to 60 beats min^{-1} at 23°C, while the rabbits' heart rates dropped from 400 to 176 beats min^{-1}. Therefore, the alligator's longer and steadier heartbeat in the face of cold temperatures ensured better coordination of the electrical activity preventing irregular heartbeats.

In short, the alligator's heart protects itself from low body temperatures by keeping robust electrical conduction so that it is unaffected by falls in temperature that would prove fatal for other species. The results underscore the trade-off between species that are restricted to lower heart rates but can function over a range of temperatures, such as the alligator, and species that can sustain a high heart rate but are vulnerable to low temperatures. The results also provide new insights into physiological adaptations of reptile hearts that allow them to maintain fluctuating body temperatures: a slow, steady tick for a cool heart.

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Herndon, C., Astley, H. C., Owerkowicz, T. and Fenton, F. H. (2021). Defibrillate you later, alligator: Q10 scaling and refractoriness keeps alligators from fibrillation. *Integr. Org. Biol.* obaa047. doi:10.1093/iob/obaa047

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Zebrafish can't take the heat around 'safer' BPAs

Pollution is, unfortunately, a permanent fixture of many environments. One particularly concerning pollutant is bisphenol A (BPA), a chemical that leaches out of plastic and can harm animals by mimicking the sex hormone oestrogen. Fortunately, BPA is being replaced increasingly in our plastics by supposedly 'safer' alternatives, such as bisphenol S (BPS) and bisphenol F (BPF). However, there are concerns that bisphenol pollutants can affect how animals respond to changing environments. How pollution and climate change – arguably humanity's two greatest legacies on Earth – interact remains a mystery. Nicholas Wu and Frank Seebacher from The University of Sydney, Australia, set out to understand how exposure to these common, physiology-modifying pollutants affected how fish performed when they turned up the heat.

Wu and Seebacher exposed zebrafish to an environmentally relevant level of BPA (30 $\mu g\ l^{-1}$), or one of its alternatives, in chilly (18ºC) and warm (28ºC) water. Then they checked how fast the fish could swim at both temperatures to get a sense of how the bisphenols interfered with the fish's ability to perform in changing environments. The BPA-exposed fish swam fine, but the fish exposed to the other bisphenols couldn't keep up: BPF and BPS tanked the top speed that all of the fish could attain. In short, it turned out that the 'safer' BPAs are not always less toxic.

To investigate how BPF or BPS exposure slowed the fish down, the researchers turned their attention to two metabolic enzymes linked to performance: citrate synthase and lactate dehydrogenase. Citrate synthase exists in the mitochondria, which provide power to all tissues. Animals with more citrate synthase have more mitochondria and more metabolic power for exercise. Lactate dehydrogenase is also involved in exercise and sometimes in adaptation to warm or cold environments

How the bisphenols affected citrate synthase activity depended on both a fish's previous experience (if they were warm or cold when exposed to the chemicals) and the temperate at which they did their swim test, highlighting how pollution could hinder fish coping with the temperature fluctuations that occur naturally in the environment. Wu and Seebacher found that when the fish were swimming in warm (28ºC) water, BPA increased their citrate synthase activity, but the enzyme was less active in the fish that had been transferred from cold to hot water. When the duo compared the effects of BPF on the enzyme, they found the opposite: higher citrate synthase activity in the fish from cold water and lower in the warm ones. In contrast, BPS reduced the enzyme's activity regardless of temperature. As the researchers had found when testing the fish's swimming abilities, BPS and BPF were just as, if not more, disruptive than the compound they were created to replace, despite their 'less toxic' reputation.

The bisphenols also impacted lactate dehydrogenase activity, but the effect depended somewhat on temperature. BPA decreased lactate dehydrogenase activity in cold fish, but the plastic increased the enzyme's activity in the warm fish, following a similar pattern as citrate synthase. BPF and BPS exposure reduced lactate dehydrogenase activities in all cases except for the fish from and swimming in warm water after a BPF exposure, which saw an increase in lactate dehydrogenase activity instead. Just like with citrate synthase, BPS and BPF were just as, if not more, metabolically disruptive than the infamous BPA.

Pollutants such as BPA and its replacements are common in many habitats and affect many physiological processes, such as metabolism, which are important for dealing with environmental challenges. Though fish exposed to bisphenols can still obtain the same top speed, they have a lot of idiosyncratic differences under the hood that limit their ability to rise to thermal challenges and beat the heat of climate change.

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