

OUTSIDE JEB

Buzzing bees grab and go while leisurely hoverflies lunch



Take a walk around the Scottish Highlands in summer and you'll be wowed by the wildflowers that light up the landscape. Bees and hoverflies are also attracted by the flowers, to collect pollen. But these two groups of buzzing insects use different behaviours to get to this protein-rich food source. Bees often buzz the pollen out of the flower, by clamping their mouthparts onto the anther and speedily shaking it with their vibrations. Buzzing pollination has evolved 40 times in over half of the 20,000 bee species, yet just one species of hoverfly is known to buzz flowers; instead, the rest silently and slowly rub the pollen off with their legs and body.

Mario Vallejo-Marin and Gillian Vallejo are no strangers to Scotland. Working at the University of Stirling and at Natural Power, Stirling, UK, they are situated right at the gateway to the Highlands and are fascinated by pollinating insects. They were curious to know why bees and hoverflies go about it in different ways.

First, the duo wondered whether hoverflies are incapable of producing large enough vibrations to shake off the pollen, so they set out to measure the bodily accelerations of bees and hoverflies during their defensive buzzes. Greater acceleration means more vigorous buzzing and more pollen. But how to measure the vibrations of a tiny insect? Use an even tinier sensor. Vallejo-Marin and Vallejo used an accelerometer

weighing just 0.2 g, to measure the vibrations. They collected bees and hoverflies from around Scotland and returned to the lab to identify the insects. They then held the insects gently against the exquisitely sensitive apparatus, just firmly enough to cause the insects to buzz with alarm and vibrate the sensor. Overall, the duo tested 299 insects, split evenly between bees and hoverflies, and measured over 4000 buzzes.

Surprisingly, the hoverflies were just as capable of producing flower-shaking vibrations as the bees. Vallejo-Marin and Vallejo found that larger insects produced larger and louder buzzes, but the buzzing produced by similarly sized bees and hoverflies were otherwise indistinguishable. They should both be able to buzz out pollen.

But buzzing a sensor is one thing; Vallejo-Marin and Vallejo wanted to see for themselves whether the hoverflies' defensive buzzes were capable of releasing pollen from flowers. They repeated the previous experiment, but this time using a natural sensor – a flower – because buzzing against an anther should cause it to release pollen. While pressing the buzzing insects against the anthers of two plant species, the researchers collected the pollen that tumbled out, and in most cases the bees and hoverflies released over 2000 grains of pollen each time: a veritable feast.

Yet despite their apparent ability, hoverflies for the most part decline to use buzzing to collect pollen. The researchers have ideas of why this might be. A big clue lies in what bees and hoverflies actually do with the pollen: the hoverflies eat it themselves, sometimes even tucking in while they're still at the flower, but the bees are there for take-away, rushing the pollen home to their larvae. Buzzing quickly exfoliates large amounts of pollen, which is a lot more effective, but more energetically expensive, than the hoverflies' relaxed and economical approach. With hungry mouths to feed, buzzing bees are certainly busier.

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Socialites boosted but loners set back in a crowd



People's responses to social situations varies considerably according to their personality type. We span the spectrum from loners to social butterflies and everything in between. And the same is true of other animals. Depending on their lifestyle, they may either be stressed or fulfilled by socializing. Megan Currier and colleagues from Widener University in Pennsylvania, USA, aimed to better understand this idea in wild animals by measuring the energy costs associated with movement in cooperative, socialite fish or antagonistic, loner fish.

Currier and colleagues suspected that the high energy investment needed to achieve movement may push some animals to maximize their efficiency through the generations. Using a fish species that is social (bluegill sunfish *Lepomis macrochirus*) versus a competitive fish with strong social hierarchies that can be found swimming alone (rainbow trout *Oncorhynchus mykiss*), the researchers set about figuring out how these animals maximize efficiency as they move through their habitat. Individuals could use what is known as 'entrainment

swimming', in which they essentially hitch a free ride by swimming in the turbulence kicked up by obstacles such as rocks or chunks of wood in the flow. Alternatively, the fishes could take advantage of the benefits of swimming in schools, when the leader of a group essentially generates comparable turbulence through the swishing of its tail, which fishy followers can exploit to reduce their own energetic costs when moving.

To test these ideas, Currier and colleagues employed a swim tunnel, also known as a fish treadmill. This contraption allowed the researchers to pump up the water flow to test how the fish optimize their swimming efficiency, by measuring the amount of energy they consumed and the rate of tail swishing. For the social bluegill sunfish, they compared the swimming efficiency of groups of three fish with that of individual swimmers, at 4 different swim speeds (ranging from 25.5 to 51 cm s⁻¹ for the 17 cm long fish). For the competitive rainbow trout, they examined the swimming efficiency of individuals versus 4 group sizes (composed of 2–8 fish), testing the fish's swimming efficiency with and without a cylindrical obstacle in the flow – for the fish to shelter behind – at a swim speed of 63 cm s⁻¹ for the 21 cm long fish. The researchers suspected that fish would use whichever strategy (either schooling or entrainment swimming) that best suited their lifestyle.

And they were absolutely right. The more cooperative and social sunfish greatly reduced both the energy consumed and their rate of tail swishing when they could do it together in a school, rather than alone, regardless of how fast the flow pumped. For the more competitive and reclusive trout, group swimming actually increased their energy use when moving around, instead of making their movement more efficient, possibly because of the stress induced by the social setting. On the flip side, rainbow trout reduced their movement costs by sheltering behind a static obstacle in the flow, taking up positions in the eddies generated by the water's flow around the obstacle. This strategy allowed the trout to reduce both their energy consumption and rate of tail swishing.

Studies like this highlight the importance of taking an animal's lifestyle into account when speculating how it will respond to animate objects (like its

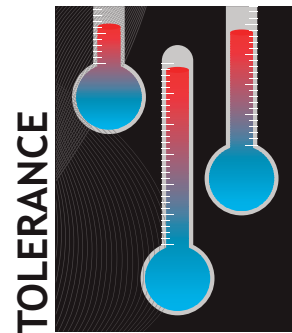
buddies) and inanimate objects (think rocks) in its environment. Like humans, some animals will be more or less social naturally and, as such, will be more or less inclined to use sociality to enhance their own efficiency. Social distancing in the ongoing COVID-19 pandemic is becoming a prolonged way of life and this study shows that keeping your distance likely impacts some individuals (like the social butterflies) more than others (such as the loners). So, some species achieve more together, but being in a crowd can set others back.

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Currier, M., Rouse, J. and Coughlin, D. J. (2020). Group swimming behaviour and energetics in bluegill *Lepomis macrochirus* and rainbow trout *Oncorhynchus mykiss*. *J. Fish Biol.* doi:10.1111/jfb.14641

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Climate change may push zebrafish to the brink



For species living near their thermal limits – the highest temperatures an animal can tolerate – climate change threatens to push them over the edge. Although zebrafish seem to be everywhere, with hundreds of investigators experimenting on different lab strains, this species may be in trouble in the wild. Zebrafish are a tropical species that live a couple of degrees below the highest temperatures at which they can survive. They live in shallow freshwater that heats up quickly, in India, Bangladesh and Nepal. Climate change scenarios predict that maximum temperatures in India are expected to surpass 44°C by 2100, leaving it unclear whether zebrafish will be able to adapt to these new conditions or if they will perish. With this chilling possibility in mind, Rachael Morgan and a team of colleagues from the Norwegian University

of Science and Technology in Trondheim set out to determine whether zebrafish can evolve heat tolerance fast enough to help them survive as temperatures soar.

Receiving zebrafish captured by local fishermen in West Bengal, India, the team allowed the new arrivals to mate back in Norway. They then placed the progeny in a tank and slowly increased the water temperature until the fish lost their balance to identify the highest temperature that each individual could cope with. Next, the team allowed the fish that could stand the heat to breed together, while allowing the least resilient fish to breed with others like themselves. Repeating the process over a total of six generations, the scientists tried to rear a hardy group that were bred to withstand high temperatures and a feebler population that could only cope with cooler conditions. The team also placed a group of the hardy zebrafish in warmer water for 2 weeks before measuring the highest temperature that each fish could tolerate to find out whether these robust animals were capable of adapting to even hotter conditions, which might further increase their heat tolerance and help them deal better with a heatwave.

Having bred at least 20,000 fish in the Herculean series of experiments, Morgan and colleagues found that zebrafish are only able to increase their thermal tolerance at a rate of 0.04°C per generation. In addition, the team found that as zebrafish evolved to endure higher temperatures, they reduced their ability to increase their heat tolerance to the same extent, using physiological changes to deal with thermal stress. These results suggest there is a limit to how much zebrafish can increase their heat tolerance.

The main question is whether the fish can adapt fast enough to keep up with the unprecedented rates of climate change that we are currently seeing and the researchers conclude it is very unlikely. Like many of the things that we have learned from this model species, these zebrafish may be warning us of what is to come for other tropical species living near their upper thermal limits.

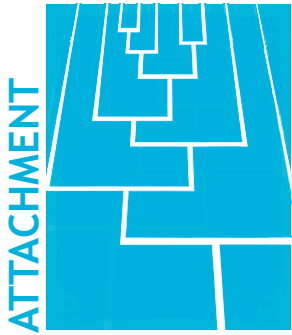
doi:10.1242/jeb.235192

Morgan, R., Finnøen, M. H., Jensen, H., Pélabon, C. and Jutfelt, F. (2020). Low potential for evolutionary rescue from climate change in a

tropical fish. *Proc. Natl. Acad. Sci. USA* **117**, 33365–33372. doi:10.1073/pnas.2011419117

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Tool-shaped ladybird hairs guarded by protein



Insects are champions of climbing even on the smoothest of surfaces thanks to their sticky footpads. The pads, found on different parts of their legs, can be smooth or hairy but both types allow the insects to attach equally well via a thin film of a liquid they secrete. Looking closely at the hairy pads, scientists have identified four distinct designs of hair tip, resembling spatulas, discs, lances or needles. But how do these tool-shaped hairs develop their different forms? Having previously investigated the hairs on the feet of fruit flies (*Drosophila*) and found that fibres of the protein actin form a scaffolding that contributes to their spatula-shaped hair tips, Ken-ichi Kimura and Naoe Hosoda, extended their research to the sticky structures on ladybird (*Harmonia axyridis*) limbs.

After collecting samples of legs from adult ladybirds and youngsters at specific life stages after moulting, the researchers examined them under a microscope. The resulting images revealed all four hair tip designs in the sticky attachment pads of each male's leg, while the female ladybird footpads contained only three, missing the disc-shaped hairs. The duo also pinpointed the location of each type of hair tip on the male footpads; finding spatula and lance-shaped hairs arranged around the perimeter of the pad, disc-shaped hairs located at the back of front-leg pads and the front of middle-leg pads, and needle-shaped hairs populating the remaining surface of the pads. Further imaging of the hairs also revealed that the hair shaft is hollow and surrounded by a

socket located on the surface of the insects' skin.

Next, the Japanese researchers focused on male ladybirds, setting out to identify how actin contributes to the development of each disc and needle-shaped hair. Using a dye that specifically targets actin, Kimura and Hosoda observed that 12 hours after moulting the footpads initially appeared flat, like terraces, with the hair sockets, hair shafts and finally hair tips progressively forming over the next 30 hours. The team was also curious to investigate the structure of the hairs beneath the insects' skin, suspecting that they might be linked to neurons, as previously reported for some fruit fly hairs. This time using a dye that reveals neurons, the scientists found that there are in fact two types of hair: one associated with a neuron, which is thought to act as a sensor and give feedback to the insects as they move, and another without a neuron.

Knowing that actin is a key player in the formation of the tip structures of *Drosophila* sticky hairs, Kimura and Hosoda wondered whether the protein was also significant in the growth and development of the ladybird's attachment hairs. Comparing the growth of normal hairs with that of hairs that had been injected with a drug that prevents actin molecules from assembling and forming long chains, the duo revealed that in both cases there were bundles of the protein in the hair shaft. However, while actin bundles branched out to form a scaffold at the hair tip in untreated ladybirds, in the insects that had been injected with the interfering drug, the scaffolding bundles failed to assemble, leading to significant malformations of the hair tips.

Fruit flies and ladybirds both depend on the same mechanisms to produce the different tip designs that help insects hold on tight. Now that the authors have verified the importance of the actin scaffolding, they are investigating new avenues, fusing developmental biology and biomimetics in search of the ultimate ladybird-inspired adhesive.

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Kimura, K. and Hosoda, N. (2021). Crucial role of framework with cytoskeletal actin filaments for shaping microstructure of footpad setae in the ladybird beetle, *Harmonia axyridis*. *Arthropod. Struct. Dev.* **60**, 100998. doi:10.1016/j.asd.2020.100998

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Freshwater snails' stinky environment could kill their babies



Growing up, an animal's environment can influence its personality. But researchers also want to know whether the parents' environment can influence the personalities of their offspring too. Recent work by Juliette Tariel and colleagues from the Université Claude Bernard in Lyon, France, suggests that this is the case for the offspring of freshwater snails (*Physa acuta*). An animal's response to predators is just one behaviour that could be affected by the parents' environment. So Tariel and colleagues wanted to see how snails would respond to a predator, and whether the youngsters' responses to a predator varied – from relaxed to terrified – if their parents were familiar with the predator's smell.

The researchers took adult snails from the river Rhône in Lyon back to the lab. They then put the snails into plastic containers filled with either freshwater or water mixed with the smell of the snail's predator – crayfish – and allowed the snails to breed. Finally, the team left some of the offspring in their original watery homes or transferred them to a container with the opposite-scented environment, predicting that offspring of parents familiar with the crayfish scent would respond faster to the predator and that there would be more variation in their individual responses. To test this, the researchers put an adult snail into a container filled with crayfish water and counted how many seconds it took for the snail to escape and crawl out of the water.

Surprisingly, Tariel and colleagues found that the parents' environments didn't affect the variation in behaviour of their offspring. All of the youngsters whose parents that had lived with the stink of predators escaped within similar time frames.

In contrast, growing up with the smell of fear did affect the youngsters' behaviour. Individual snails often responded differently from their brothers and sisters; there was much more variation in their behaviour. The difference in the baby snails' responses might be due to differences in how they use energy while developing. For example, some snails may invest energy into growing thicker shells for defence rather than building muscle for a sprint escape.

Another surprise was that the snails produced by parents that bred in the crayfish-ridden water tended to escape more slowly when presented with a predatory crayfish; if they tried to escape at all. The snails' slow escape might be because the snails are familiar with the odour of crayfish. The scent of crayfish usually warns snails when a predator is at large, but if the snails constantly smell crayfish without encountering the genuine threat, they could well stop responding to the scent as they appear to have nothing to fear.

While this is the opposite of the researchers' original prediction, the discovery that crayfish odour carried less dread for the offspring of parents living

with the scent of fear suggests that the environment of parents can affect how their babies respond to changes in their own environments. In the case of these freshwater snails, where their parents grew up could be a matter of life and death, if the youngsters have lost their edge and don't know when to make a speedy getaway.

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Tariel, J., Plenet, S. and Luquet, E. (2020). How do developmental and parental exposures to predation affect personality and immediate behavioural plasticity in the snail *Physa acuta*? *Proc. R. Soc. B* **287**, 20201761. doi:10.1098/rspb.2020.1761

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