

OUTSIDE JEB

A mucous house built for feeding



Some of the world's weirdest and most fascinating creatures live in the deep sea, but frustratingly for marine biologists, these creatures are also some of the hardest to study. The remoteness, darkness, and high pressures of the deep make human observation difficult, and many deep-sea animals are too fragile to study in captivity. This means most people have never heard of these marvelous creatures, let alone understand how they make their living. Giant larvaceans are one of these amazing, but largely unknown, animals. The worm-like invertebrates float around the deep sea encased in a pair of large (up to 1 m diameter) mucous houses, with one tucked within the other. The outer house is thought to be a protective structure, while the smaller inner house is used for filter feeding on tiny plankton. However, the details of the larvacean filtering mechanism have been only superficially understood because of the immense technical limitations of studying a large and fragile ball of mucus drifting through the deep ocean.

A new study, led by Kakani Katija at the Monterey Bay Aquarium Research Institute, USA, used a sophisticated laser imaging system attached to a remotely operated vehicle to study wild giant larvaceans and their mucous homes at depths up to 400 m. By shining a laser at the animals while the robot carefully maneuvered around them, the edges, folds and other intricacies of the mucous house could be illuminated and photographed. Then, back on shore, the team used these images to generate a complete 3D model of the filter feeding mechanism. At sea, the team also used their underwater robot to release small amounts of fluorescent dye into the water, which allowed them to observe flow patterns throughout the mucous houses without otherwise disturbing the animals.

The team discovered that the larvacean house is an amazingly complex structure, especially considering it's made entirely from mucus and may be discarded and replaced daily. Sea water, pumped by the beating tail of the larvacean, enters the inner house via two long tubes that extend to the periphery of the outer house, passing through a protective pre-filter on the way. The flowing water is then divided into a symmetrical pair of foodconcentrating filters, which allow water to exit while food particles are trapped and then passed directly to the larvacean's mouth. A complex series of valves, extra chambers and connecting threads further regulate water flow and internal pressure, enabling the mucous house to remain properly inflated.

Thanks to this cutting-edge imaging technology, the structure and function of the inner mucous house is now well understood. Getting good images of the thin mucous walls of the larger, outer larvacean house remains a challenge, however, and so the details of its structure and function remain unknown. One possibility is that the mucus serves as a coarse filter that prevents large particles from clogging the delicate inner feeding apparatus. The outer house may also deter predators by acting as a physical barrier, or even by functioning as a cloaking device that muffles the turbulence produced within the inner house during filter feeding. Finding the answer will require further advances in deep-sea robotics, laser-assisted video recording, and other technologies, but given the recent achievements of Katija and her crew, finding answers to these sorts of questions finally seems within the realm of possibility.

doi:10.1242/jeb.214544

Katija, K., Troni, G., Daniels, J., Lance, K., Sherlock, R. E., Sherman, A. D. and Robison, B. H. (2020). Revealing enigmatic mucus structures in the deep sea using DeepPIV. *Nature* 583, 78–82. doi:10.1038/s41586-020-2345-2

> Andy Turko (0000-0002-6330-5798) University of Windsor and McMaster University ajturko@gmail.com

Bats pay the price for crying out loud



Bats are often depicted as silent and stealthy hunters of the night, but they are also some of the loudest animals on the planet, capable of blasting out highfrequency calls with the same sound pressure as a jet engine. Many bat species use these high intensity shouts to echolocate their prey during foraging flights. Until recently, it was thought that the energetic cost of echolocation during flight was negligible for bats, since there was seemingly no difference in the flight costs for echolocating and nonecholocating bats. Yet, bats in the wild quite often hunt in groups and each bat must compete with a chorus of other calls to echolocate effectively, but at what cost? A new study by researchers from the Leibniz Institute for Zoo and Wildlife Research in Berlin, Germany, shows that singing for your supper can be very costly indeed in noisy environments.

To test how these aerial shouting matches affect the energy consumption of bats during flight, the researchers flew Nathusius' pipistrelle bats (*Pipistrellus nathusii*) in a wind tunnel and measured their metabolic rates while exposing them

Outside JEB reports on the most exciting developments in experimental biology. The articles are written by a team of active research scientists highlighting the papers that JEB readers can't afford to miss.

to different levels of background noise. To create a low-noise flight environment, the researchers simply used the ambient noise of the wind tunnel (69 dB): however, to achieve a high-noise flight environment, they placed loudspeakers under the wind tunnel playing 'white noise' at 109 dB and placed a microphone upstream of the bat in the tunnel to record the intensity of its echolocation calls. Finally, in order to measure the energetic expenditure of the bats during flight, the researchers started each experimental flight by injecting the bats with a carbon-13 isotope, which the animals would exhale as CO₂. By taking breath samples from the bats before and after each flight, the team measured the difference in the ¹³C concentrations to estimate how much CO_2 had been produced as a reliable proxy for the animal's metabolic rate.

Unsurprisingly, all of the bats' calls were much louder (128 dB) when competing with the high background noise, in comparison with their calls when the ambient noise was low (113 dB). In fact, the high-noise wails were actually 30 times louder than the low-noise whispers, thanks to the exponential decibel scale. More importantly, while the team found that the costs of echolocating in low-noise environments are negligible, averaging just 0.8% of the flight costs, echolocating in loud background noise caused the bats to drastically boost their own calls, with the price of echolocation skyrocketing to almost 22% of the total flight costs. This completely overturns previous assumptions about echolocation energetics and the team calculated that the cost of calls exceeding 130 dB may be even greater than the cost of flight itself.

In the context of wild bats, the team concluded that during a single night of foraging in a high-noise environment, such as hunting in the vicinity of other bats, they would need to eat an additional 0.5 g of fresh insects to balance out the calling costs – which is a sizeable additional snack for a 7 g bat. It's not all bad news though as the team also found that the higher intensity calls allowed bats to find smaller insects at greater distances, albeit with diminishing returns for the most costly calls. On a final note, the team concluded that, interestingly, it's not just bats that cap their vocal intensity at \sim 130 dB, as the peak intensity for many other mammals, birds and amphibians is also in the 130–140 dB range, suggesting

that it may be energetic constraints, rather than body size, that limit the ability of animals to shout, scream and squawk.

doi:10.1242/jeb.214528

Currie, S. E., Boonman, A., Troxell, S., Yovel, Y. and Voigt, C. C. (2020). Echolocation at high intensity imposes metabolic costs on flying bats. *Nature Ecol. Evol.* 4, 1174–1177. doi:10.1038/ s41559-020-1249-8

> Alex Evans (0000-0002-5655-012X) University of Leeds alexevans91@gmail.com

Andean condors take cheap flights



The largest living soaring birds are reluctant flappers. Wing flapping costs calories, and if the next meal is hundreds of kilometres away, saving energy is most important. The Andean condor (Vultur gryphus), a scavenger weighing up to 16 kg, is a soaring specialist. These birds are known to avoid flapping their wings until it's absolutely necessary and rely instead on external air currents to subsidise their flight costs. Given this reliance on air currents, learning how condors interact with their surroundings might reveal the limits to their aerial athleticism and may even offer a glimpse into the past, when impossibly giant birds and pterosaurs roamed the sky.

Hannah Williams, Emily Shepherd and colleagues from Swansea University, UK, together with Sergio Lambertucci and colleagues from the *Universidad Nacional del Comahue*, Argentina, decided to take a closer look at condors flying in the sky. This goes beyond casual birdwatching – how do you observe an animal cruising 1 km high for hours on end? Williams and Shepherd used remote data logging devices. They caught eight juvenile condors in a mountainous region of Argentina and tagged them with the devices before quickly releasing the animals. The loggers measured the birds' location and motion, as well as the ambient pressure and temperature around them, and were pre-programmed to drop off the birds while they were roosting. Williams and Shepherd specifically chose to study juvenile birds as they are more likely to roost in accessible locations – handy for retrieving the loggers later.

Incredibly, the condors covered a lot of ground with their wearable tech, with one bird travelling over 300 km in a single day. In addition, they flapped for only 1% of their total flight time of 235 h, which is a record low for powered flight in any bird. One of the condors even went for 5 h in the air, covering 170 km without flapping once. This is a very cheap way to get around, as riding on air currents requires 15 times less energy than powered flapping for the equivalent time. On average, the birds resorted to flapping for only 4 mins per day, with three of these minutes spent taking off from the ground. This behaviour didn't even change much with weather conditions, telling us that the birds' athletic abilities aren't strictly restricted by the environment around them. Instead, the amount of time they spent flapping was linked to the effort required to take off. The energy used during 3 min of take-off is equivalent to 50 min of soaring, so it's critical for the condors' calorific budget that they don't land unnecessarily.

The other incidences when the condors resorted to flapping usually occurred as the birds moved between thermal updraughts, particularly when the thermals were weak, or when the birds were at low altitude. Given the risks of grounding, when they are most vulnerable to predation by opportunistic mammals, the condors invest precious energy flapping their wings to get to the next energy-saving thermal.

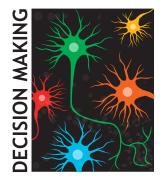
The researchers conclude that for Andean condors, landing is risky and take-off is expensive, but flight is cheap. These heavy birds exploit their environment to their full potential, allowing them to travel colossal distances on a tight budget.

doi:10.1242/jeb.214569

Williams, H. J., Shepard, E. L. C., Holton, M. D., Alarcón, P. A. E., Wilson, R. P. and Lambertucci, S. A. (2020). Physical limits of flight performance in the heaviest soaring bird. *Proc. Natl. Acad. Sci. USA* **117**, 17884-17890. doi:10.1073/pnas.1907360117

Christopher Basu (0000-0002-2357-9516) Imperial College London c.basu@ic.ac.uk

Wasps choose their battles wisely



Deciding when to stand your ground and when to retreat can be a life-saving skill. One way to decide whether to pursue peace or combat is to know something about your rival and your own chances of victory. If you don't think you'll have the upper hand, it's better to conserve energy. This deliberation requires abstract social predictions – calculations that some people believe emerge only in larger brains. In a paper wasp species, however, previous work suggests that the wasps identify peers by unique facial markings to make social inferences about each other, suggesting that brain size might not be a limiting factor for social intelligence.

Recently, a team led by Elizabeth Tibbetts at the University of Michigan, USA, set out to test whether wasps inform social interactions just from watching others without any direct contact. Specifically, they wanted to learn if wasp aggression is predicted by the wasps' observations. They set up a battle arena - a box where two queen wasps were placed in close quarters until they eventually came to blows. Meanwhile, other queens observed from outside through a clear divider. As the spectators watched the fight, the researchers monitored them and the fighters. The scientists then rated each combatant on an aggression scale based on how often the queens bit, grappled and attempted to sting, in addition to keeping track of how long the spectators observed.

Once the first round of combat was complete, one of the spectators was

placed in its own battle arena with one of the queens that it had just watched fight. If the queen had been the aggressor in the previous fight, the spectator was less aggressive, while the spectator was more aggressive when presented with the loser from the preceding altercation. In order to determine whether the wasps are indeed making social inferences and adjusting their behaviour if they expect to win or lose, the team needed to rule out two alternative explanations: that watching a fight influences wasps to be more aggressive, like watching a violent movie, or that winners keep winning while losers continue to lose.

To test whether simply watching a fight primed the observer to be more aggressive, the spectator was matched against a stranger. If simply being a bystander at a previous contest increased the wasps' aggression, the team reasoned that combatants who had previously been spectators would pick fights with any wasp that they were pitted against. However, some of the wasps were aggressive when set against an opponent. while others were not; just watching a fight did not inspire violence in the wasps. To test whether winning or losing previous fights dictates the outcome of future fights, the team set up two consecutive battles with different challengers. If the losers continued to suffer more defeats and winners vanquished their opponents again and again, fighting performance might not be a choice based on social inferences. However, the researchers did not see any relationship between how dominant a wasp was in one fight compared to the next; winners weren't always winners. In short, paper wasps learn about their peers and adapt their behavior in response to what they see.

What is more remarkable is that they pull off this feat with such small nervous systems. Even wasps decide when they can safely fight a foe with a weaker track record, or walk away to save energy and fight another day.

doi:10.1242/jeb.214551

Ellen Lesser (0000-0001-7929-0503) University of Washington elesser@uw.edu

A beach festival isn't a sound idea to fish



Music is a common language across many cultures and is often celebrated with large festivals that can attract thousands of people in just a single day. For festivalgoers, the relentless melodies and rhythms can be exhilarating, but for nearby neighbours, it can be an intrusive din that can leave some cantankerous and deprived of sleep. The booming noise can also disrupt local wildlife. In 2019, the annual 3-day Ultra Music Festival in Miami, Florida, was located on an open beach, for the first time. But the proximity to the water worried scientists; the coast is home to an abundance of marine life, including several fish species, dolphins and manatees – all of which have the ability to hear and are already disturbed by noisy human activity. Curious about the effects of the festival on sea life, Maria Cartolano and her colleagues from the University of Miami, USA, tested the response of fish to the sound of the Ultra Festival.

Three weeks before Ultra began, the team set up outdoor fish tanks at the University of Miami's aquaculture facility, located down the road from the festival's location. They then settled Gulf toadfish (Opsanus *beta*) – a local species that was likely to be affected by the festival's acoustics - into the aquaria, to find out how they would react to the wall of sound that was about to hit them. Next, Cartolano and colleagues took blood samples from the fish to measure their baseline (relaxed) levels of cortisol, a stress hormone. If the festival were to perturb the fish, then the biologists expected these cortisol levels to increase. The team then set up a sound meter on the same beach as the festival, to measure the volume of the music in the air, and hydrophones, to measure the underwater sound levels of the beach and in the fish tanks, focusing on the low

Tibbetts, E.A., Wong, E. and Bonello, S. (2020). Wasps use social eavesdropping to learn about individual rivals. *Curr. Biol.* **30**, 3007–3010. doi:10.1016/j.cub.2020.05.053

sound frequencies between 100 and 1000 Hz that the fish hear.

During Ultra, the sound meter detected that air noise levels rose almost 15-20%, which were matched by modest increases in the underwater lowfrequency noise -3% off the beach and 7% in the tanks. Despite the small increases in underwater sound, the fish's blood cortisol levels rose by an average of 400%. This abrupt increase in cortisol demonstrated that the loud music produced by the festival had severely stressed the fish. More worrying, however, was that some of the fish were affected more considerably than others. Indeed, 10 h after the first day of the festival, when the fish were returned inside the lab where the festival

music was inaudible, the health of eight of the fish seemed to have deteriorated substantially and the scientists recorded an enormous 23-fold increase in blood cortisol levels, relative to the cortisol levels of the remaining healthy-looking fish. In fact, the acoustic stress produced by Ultra increased the toadfish's cortisol to levels that affect their social interactions, communication and their ability to avoid predators.

This study provides useful insight into how sound pollution from human activities can alter noise levels in the water, which can profoundly change the biology of marine organisms. In particular, species that use sounds to communicate and socialize could be especially affected by loud acoustics from festivals, potentially harming the animals' abilities to reproduce or even survive. Clearly, more research is need to fully determine how human noise levels are impacting coastal marine life, but it sounds as if relocating the Ultra festival away from the beach might be a better option for our marine neighbours next time around.

doi:10.1242/jeb.214536

Cartolano, M. C., Berenshtein, I., Heuer, R. M., Pasparakis, C., Rider, M., Hammerschlag, N., Paris, C. B., Grosell and M. and McDonald, M. D. (2020). Impacts of a local music festival on fish stress hormone levels and the adjacent underwater soundscape. *Environ. Pollut.* 265, 114925. doi:10.1016/j.envpol.2020.114925

> Ilan Ruhr (0000-0001-9243-7055) The University of Manchester ilan.ruhr@manchester.ac.uk