Inside JEB is a twice monthly feature, which 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.

Inside JEB

FROG SWIMMERS KICK AND ROW

If you're a breaststroke swimmer, you might think you know all about frog swimming. But take a closer look and you'll find that there's a lot more going on. For a start, many frog swimmers do more than simply push off with their legs; some row with their paddle-like feet by rotating their ankles to push against the water. But how much of a contribution do leg-pushes and foot-paddles make as a frog moves through its complex aquatic world? Curious to find out more, Christopher Richards decided to film swimming frogs to determine exactly how they move and then built a mathematical frog to dissect out the leg and foot-paddles' contributions to frog propulsion (p. 3181).

Richards explains that he chose to work with African clawed frogs, *Xenopus laevis*, because they are pure swimmers, spending all of their lives in water. 'Their legs are super muscular and they are very powerful animals,' says Richards, adding that handling the slimy creatures as they struggled to get free while he superglued markers to their legs and body was quite challenging. And getting the frogs to swim was even trickier; 'they become habituated to the tank and are reluctant to swim,' he explains. The frogs seemed to respond best when he turned on the lights to startle them.

After weeks of patiently waiting for the frogs to swim and analysing their style, Richards set about developing a mathematical model of the frogs' swimming through water so that he could calculate the contributions of their pushing legs and rotating feet to propulsion. He admits that getting the model to work was difficult, as he had to model the behaviour of each joint in the leg and ankle as well as the interaction between the animal's foot-paddles and the water it dragged along. But Richards eventually found a piece of software that could solve the complex equations of motion and was able to begin modelling the animal's behaviour.

Simulating frogs swimming normally, as well as only kicking their legs or only rowing with their feet, Richards discovered that the animals rely far more on their feet rotating than on their leg pushes. The mathematical frogs only reached a top speed of 0.38 m s^{-1} when powered by leg pushes alone but rocketed to 0.54 m s^{-1} when powered entirely by foot rowing. Analysing the simulations based on real swimming strokes, Richards realised that the frogs depend on their leg pushes at the beginning of a stroke but that their rotating feet take over later in the stroke, providing the majority of the thrust. Richards also noticed that the frogs decelerated more towards the end of a stroke when kicking with their legs alone because of water resistance caused by their feet sticking out. By rotating their feet at the end of the stroke, the foot-rowing frogs were able to reduce the resistance significantly.

Satisfied that his mathematical frog is working as well as the real thing, Richards is keen to use his model to find out how other frogs propel themselves through water, as well as to understand how the animal's powerful leg muscles operate over multiple joints to generate the frog's hefty swimming kick.

10.1242/jeb.024570

Richards, C. T. (2008). The kinematic determinants of anuran swimming performance: an inverse and forward dynamics approach. *J. Exp. Biol.* **211** 3181-3194.

BATS PICK UP RUSTLES AGAINST HUMAN NOISE

Listening for faint rustling noises made by tasty beetles on a quiet day is simple for bats hunting with their exquisitely sensitive hearing. So try imagining what it must be like trying to locate rustling treats just metres from a roaring highway. It would seem to be almost impossible to pick out a centipede's footsteps as a juggernaut hurtles past; or is it? How animals that locate their prey by sound alone cope in our increasingly noisy world puzzles Björn Siemers from the Max Planck Institute for Ornithology in Germany. Siemers explains that no one had ever measured whether bats that hunt by listening for rustling insects are affected by man-made noise. However, this is a question that Siemers is frequently asked by urban planners keen to minimise our impact on local wildlife populations. Curious to know how sharp-eared bats react to loud background noise, Siemers and his colleagues Andrea Schaub and Joachim Ostwald monitored foraging bats' responses to rustling mealworms in noisy environments (p. 3174).

Working with a group of young male greater mouse-eared bats, Siemers and Schaub allowed individual bats to forage freely in a large soundproof room. Dividing

the back of the room in two, the duo provided the bats with a choice of rustling mealworm snacks in each half of the room to dine on. Over the course of several days, the bats divided their attention equally between the two halves of the room, easily locating the rustling nibbles. But how would the bats react when the team switched on a noise in one of the two dining areas?

First, the team synthesized true white noise before playing the sound in one half of the flight arena. The bats instantly avoided the unpleasant buzzing sound, spending more than 80% of their time hunting in the quiet dining area.

Next, Siemers and Schaub headed out to a local highway to record traffic sounds within 15 m of the busy road. Back in the lab the animals were less bothered by the loud traffic noise than they had been by the white noise buzz. But they still preferred hunting in the quiet dining area, only spending 38.7% of their time gathering mealworms from the traffic noise booth. However, when the bats ventured into the noisy dining area they had no obvious problems locating their rustling prey against the traffic background.

More surprisingly, when the team played a simulation of a high wind rattling reed beds, the bats seemed to find it difficult to locate their prey and preferred foraging away from the sound, even though it was a noise that they encounter naturally in their day-to-day activities. So, man-made noise does interfere with bat foraging, but less than a very high wind rattling through vegetation.

Siemers admits that it isn't yet clear how man-made noise interferes with foraging bats as they listen out for a rustling lunch, but it probably does discourage these animals from foraging close to busy road networks. Keen to find out whether noise pollution affects foraging bat's hearing, or interferes with some other aspect of the bat's behaviour, Siemers is optimistic that his work will eventually result in bat-friendly road construction guidelines that will help to protect endangered bat species from our increasingly mechanised world.

10.1242/jeb.024588

Schaub, A., Ostwald, J. and Siemers, B. M. (2008). Foraging bats avoid noise. *J. Exp. Biol.* **211,** 3174-3180.

IS AMPK THE HYPOXIA TOLERANCE MASTERSWITCH?

Some creatures are better at coping without oxygen than others. Humans keel over after just three minutes, but goldfish can keep going for months if it's cool enough. Jeff Richards explains that the key to survival is balancing your energy demands. Most creatures that survive low oxygen levels drastically reduce their energy consumption by shutting down processes such as protein synthesis and ion transport to preserve meagre energy supplies. But how do hypoxia-tolerant creatures coordinate the complex array of energy-conserving events that protect them from otherwise certain death? At the time that Richards began puzzling this question, he was working on exercise in muscles. It occurred to him that the challenges of matching energy consumption with supply in exercising muscles were similar to those faced by hypoxia tolerant animals. Knowing that a protein, AMP-activated protein kinase (AMPK), was the key coordinator in exercising muscle, Richards says 'a light went on in my head'; maybe AMPK was the master switch for hypoxia tolerance too. He decided to see if AMPK was involved in metabolic regulation in a hypoxia tolerant species: the goldfish (p. 3111).

Setting up his own lab for the first time, Richards recruited Master's student Lindsay Jibb to begin discovering whether AMPK is significant in hypoxia tolerance. But without a genome to fall back on, Jibb first had to set about cloning individual subunits from the kinase to find out which tissues produce the protein. Not surprisingly, AMPK turned up in the brain and kidney, but the duo also found it in the fish's liver. Knowing that the liver of other hypoxia-tolerant species drastically reduces metabolic activity when faced with low oxygen levels, Richards decided to focus on the role of AMPK in goldfish liver.

Next, the team decided to see how the fish's liver responded to hypoxia. The first thing that they noticed was that ATP levels in the liver dropped significantly during the first 30 min of hypoxia. Richards admits that this was surprising; one of the hallmarks of hypoxia-tolerant species is that their ATP levels remain constant, even when oxygen levels fall. However, after this initial drop, the goldfish's ATP levels stabilised.

Having found that the goldfish were able to maintain their energy supplies, even when oxygen levels were low, Richards and Jibb decided to see if the goldfish's AMPK was capable of activating other proteins. Knowing that AMPK in muscle adds a phosphate molecule to enzymes to activate them, the duo tested whether the goldfish AMPK was also capable of transferring a phosphate molecule to target proteins. Supplying liver tissue containing AMPK with radioactive ATP and a protein fragment that could only be phosphorylated by AMPK, the team found that AMPK successfully transferred a radioactive phosphate from the ATP to the protein fragment. AMPK could active key proteins involved in hypoxia tolerance.

Richards admits that this discovery was very exciting, but then Jibb suggested taking the experiments even further; could goldfish AMPK add a phosphate to a protein that must be switched off to save energy? Could AMPK phosphorylate and inactivate Elongation Factor 2, a key protein in protein synthesis? This time the team used antibodies to identify whether AMPK transferred a phosphate to Elongation Factor 2, and it did. AMPK could switch off protein synthesis.

Richards admits that this is all circumstantial evidence that AMPK is the master regulator of hypoxia tolerance in goldfish, but hopes to prove one day that AMPK is at the hub of hypoxia tolerance.

10.1242/jeb.024596

Jibb, L. A. and Richards, J. G. (2008). AMP-activated protein kinase activity during metabolic rate depression in the hypoxic goldfish, *Carassius auratus*. *J. Exp. Biol.* **211** 3111-3122.

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