Inside JEB 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

## HOW PYGMY KILLER WHALES HEAR


Whales and other cetaceans live in an almost exclusively acoustic world. Dependent on their hearing to hunt, communicate with other members of their species and interact with their environment, whales rely heavily on their auditory sense to survive. Yet we know little about these animals' acoustic processing, much less how our activities impact on them. So when two pygmy killer whales were rescued by the Mote Marine Laboratory Dolphin and Whale Hospital after stranding on a Florida beach in June 2008, Eric Montie from the University of South Carolina Beaufort, USA, and David Mann from the University of South Florida, USA, grasped the opportunity to find out more about the hearing of these elusive creatures (p. 945).

Montie recalls, 'Charlie Manire called David and said, "We've got these two stranded pygmy killer whales, are you interested in doing some hearing tests on these animals?"; they were trying to get an idea of whether or not there were any hearing deficits because you don't want to release a cetacean into the wild that has severe hearing loss or is deaf.' Given this rare chance, Montie and Mann tested the pygmy killer whales' hearing while Manire cared for them. The team measured the animals' brain electrical activity as they played a series of 14 ms beeps through the whales' lower left jaw, starting at the lowest pitch of 5 kHz up to the highest pitch of 120 kHz, at various sound pressure levels (volumes) to test the whales' hearing sensitivity. Then they measured the brain's response at several locations on the surface of the whales' heads as an auditory signal travelled through – and was processed by – different brain regions.

Although both of the animals were most sensitive to frequencies ranging from 20 to 60 kHz and could hear frequencies as high as 120 kHz, their high-frequency hearing was not as good as that of other toothed whales.

Next, the team CT-scanned the head of one of the pygmy killer whales to find out more about the animal's hearing. Reconstructing a 3D image of the whale's brain and auditory system, the team could see that structures in the pygmy killer whales' auditory system were similar to those of other toothed whales. The jawbones were hollow and packed with fat that pressed against the tympanoperiotic complex to transmit sound to the middle and inner ear for processing.

With a clear picture of the whale's hearing system and acoustic electrical activity patterns recorded from the brain at several sites across the animal's head, Montie and Mann realised that they could begin to identify which regions of the brain were involved in processing information about the whale's complex acoustic environment. Analysing the strength of the brain stem's response over the surface the whale's head, Montie and Mann conclude that the auditory nerve and two brain regions – the inferior colliculus and medial geniculate body – generate some of the electrical signals and process the complex sounds that steer whales through life.

Having tested the hearing of these two pygmy killer whales, Montie is keen to investigate the effects of man-made pollution on dolphin and whale hearing. 'I have a training in marine toxicology so I'm very interested in whether or not the pollutants that marine animals accumulate throughout their lifetime, and then transfer to their young through milk, affect how they hear,' says Montie. He explains that some man-made pollutants can affect the thyroid system, which could affect hearing development in young cetaceans, with potentially catastrophic effects for future generations.

10.1242/jeb.056788

**Montie, E. W., Manire, C. A. and Mann, D. A.** (2011). Live CT imaging of sound reception anatomy and hearing measurements in the pygmy killer whale, *Feresa attenuata*. *J. Exp. Biol.* **214**, 945-955.

## SALMON HEARTS DO NOT FOLLOW MASS SCALING LAW

Most large creatures have much lower heart rates than smaller a nimals. In fact, it is even possible to find a correlation between body size and heart rate, which has led some scientists to propose a model that could explain this phenomenon. But do all animals conform to this scaling law? Mounting evidence suggests that this may not always be the case. Timothy Clark and Tony Farrell from the University of British Columbia, Canada, point out that little is known about the scaling relationship between body mass and heart rate in fish. Curious to find out whether fish heart rate


patterns follow the same scaling trend as other vertebrates, Clark and Farrell caught nine male Chinook salmon, ranging from 2.5 to 15.9 kg, as they migrated to their spawning grounds and implanted data loggers that could record each fish's heart rate and activity patterns. Next, Clark and Farrell allowed the animals to swim freely in a holding channel for 9 days before retrieving the data loggers (p. 887).

However, when they analysed the data, there was no correlation between the fish's body size and heart rate. Instead of decreasing with size, the fish's resting heart rates were all between 30 and 43 beats $min^{-1}$. And when the duo considered heart rate data from the literature for tiny 0.02–0.05 g trout, they found that even the smaller fish only had heart rates of 50–60 beats $min^{-1}$, rather than the 1000 beats $min^{-1}$ range you would expect if they followed the same size scaling pattern as birds and mammals.

Next, the duo analysed the relationship between the fish's body size and various blood parameters, such as haemoglobin and glucose levels. They found that all of the parameters varied, but in no consistent way relative to the fish's body masses. Meanwhile, the fish's heart chamber (ventricle), spleen and heart muscle (myocardium) masses all scaled in proportion to the fish's body masses. The duo suggest that the Chinook salmon's ability to transport oxygen and their mass-specific cardiac output and cardiac power may be maintained across all body masses.

Having shown that the scaling law for heart rate does not hold in Chinook salmon, Clark and Farrell point out that fish offer great potential for examining the effects of body mass on other physiological variables, as their body masses vary to a greater degree than any other vertebrates and they account for over half of all vertebrate species.

10.1242/jeb.056796

**Clark, T. D. and Farrell, A. P.** (2011). Effects of body mass on physiological and anatomical parameters of mature salmon: evidence against a universal heart rate scaling exponent. *J. Exp. Biol.* **214**, 887-893.

## GENE EXPRESSION SHOWS NO TWO MEMORIES ARE THE SAME


Whenever a memory is formed, thousands of new connections – synapses – form in the brain, but what underlies these changes? For example, do the expression patterns of brain genes differ between memories? This is the question that Gene Robinson and his colleagues from University of Illinois Urbana-Champaign and East Tennessee University asked. Robinson explains that honeybees are an ideal species for answering this question because so many elements of their environment can be controlled with precision, and the impact of these changes on the insects' behaviour and gene expression patterns can be analysed. Curious to find out how the bee's brain gene expression patterns differ when forming two similar, but distinct, memories, Robinson and his colleagues trained groups of bees to go foraging at different times of day and in different places and then looked for differences in the insects' brain gene expression patterns (p. 979).

Robinson's student, Nicholas Naeger, travelled to Darrell Moore's lab in Tennessee where he teamed up with Byron Van Nest, Jennifer Johnson and Sam Boyd to train honeybees to visit one of two feeders. Robinson explains that flowers open and produce nectar and pollen at specific times of day and bees learn to remember this. So, the team trained one group of bees to forage at a lilac-flavoured feeder in the morning and another group of bees to forage at a lavender-flavoured feeder at a different location in the afternoon. Having successfully trained 40 bees to feed at each feeder, the team collected bees from both groups 15 min before their normal departure while they prepared to go foraging. Naeger and his colleagues also collected the trained bees when they were inactive, because the insects' activity levels and time of day can affect brain gene expression patterns and Robinson wished to account for these factors in the gene expression patterns.

Returning to Urbana-Champaign, Naeger extracted mRNA from the insects' brains and then used a microarray analysis where he could simultaneously compare the expression levels of 11,000 honeybee genes between two groups of bees. Comparing different combinations of bees (e.g. morning-trained active bees with morning-trained inactive bees or afternoon-trained active bees with afternoon-trained inactive bees), the team eventually identified 1329 genes (over 10% of the honeybee genome) that showed expression pattern changes in response to the different memories. And when Sandra Rodriguez-Zas statistically analysed the complex microarray gene expression patterns and took out genes involved in time keeping and activity, which are essential to the memory, the team narrowed down the number of genes that were differentially expressed in the two memories to 352.

Identifying groups of genes that respond differently in the two memories, Robinson highlights genes involved in synapse formation and genes involved in other forms of chemosensory behaviour. 'The specific genes are interesting, but what is more interesting is to consider the kind of memories we are looking at. It is known that changes in synapses are associated with building memories. However, both groups are building the same kind of memory – it's just for a different place and time – and we see still see differences – this hints at previously unknown forms of specificity for synapse formation in memory,' says Robinson. He adds, 'The differences are extensive, telling us that no two memories are alike when it comes to the genome.'

Ultimately Robinson hopes to identify specific categories or genes involved in memory formation, and to identify genes that are diagnostic of specific memories and their locations in the brain.

10.1242/jeb.056762

**Naeger, N. L., Van Nest, B. N., Johnson, J. N., Boyd, S. D., Southey, B. R., Rodriguez-Zas, S. L., Moore, D. and Robinson, G. E.** (2011). Neurogenomic signatures of spatiotemporal memories in time-trained forager honey bees. *J. Exp. Biol.* **214**, 979-987.

## NON-STICK HAIRS KEEP CRANEFLIES DRY


Large animals think nothing of walking through a heavy mist: water droplets simply role off their hides. However, smaller insects are at constant risk of entrapment by the sticky forces of surface tension. Craneflies, which set up home in boggy settings and riverbanks, routinely encounter damp surfaces and mist that could prove fatal, yet they shrug off droplets with ease and can even stand on water. Jolanta Watson and colleagues from James Cook University and the University of Queensland, Australia, decided to take a close look at the insect's fragile legs and wings to find out how craneflies avoid getting stuck in water (p. 915).

Photographing cranefly legs at increasing magnification, the team could see that the insect's legs are covered with water-repelling hairs: thick long (90 µm) hairs with a rough grooved surface, shorter thick curved hairs, even shorter fine hairs and the shortest hairs of all found clustered at the base of the longest thick hairs. The insect's wings are also covered in fine hairs, with 12 µm long hairs distributed evenly across the membrane and 90 µm long hairs coating the wing veins.

To find out how repellent the hairy surfaces are, the team photographed water droplets on the insect's legs and wings. They saw that instead of spreading over the insect, the droplets formed perfect spheres, characteristic of the way water is repelled by a hydrophobic surface. And when they laid a cranefly leg on water, the hairs formed tiny dimples in the surface instead of piercing it.

Finally, the team tested how the grooves on the longer hairs help the insects repel water by coating the long hairs with hydrophobic polydimethylsiloxane to fill the grooves. Poking coated and uncoated hairs into water droplets, the team could see that the coated hairs no longer repelled water and penetrated the droplets with ease, while the uncoated hairs were unable to pierce the droplets.

So craneflies avoid getting trapped in sticky water with a coating of rough hydrophobic hairs. Watson and her colleagues are keen to design cranefly-inspired water-repelling and self-cleaning surfaces.

10.1242/jeb.056770

**Hu, H.-M. S., Watson, G. S., Cribb, B. W. and Watson, J. A.** (2011). Non-wetting wings and legs of the cranefly aided by fine structures of the cuticle. *J. Exp. Biol.* **214**, 915-920.