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Inside JEB

ASIAN AND EUROPEAN HONEYBEES USE ANCESTORS' BUILDING STRATEGY



At first glance, humans no longer seem to have much in common with our ape ancestors, but what about other species that diverged at a similar time? How much do Asian honeybees (*Apis cerana*) and their European counterparts (*Apis mellifera*) have in common after 2 million years of independent evolution? Ming-Xian Yang from Rhodes University, South Africa, and Yunnan Agricultural University, China, explains that despite having completely different appearances – Asian honeybees are much smaller than European honeybees – and strategies for preventing their nests from overheating, the insects are still able to communicate through their waggle dances and can even live together harmoniously in hives. Intrigued by the bees' unexpected ability to cooperate, Yang and his colleagues Ken Tan, Sarah Radloff and H. Randall Hepburn decided to find out whether mixed colonies of Asian and European honeybees could successfully construct honeycombs together (p. 1659).

According to Yang, honeybees build comb following a specific construction strategy. 'In a dark hive there is no leader and no blueprint for them to follow. Every bee has to perform by herself and I wanted to see if one species could collaborate with another,' explains Yang. Travelling to Thailand to work with Mananya Phiancharoen from King Mongkut's University of Technology Thonburi, Yang took combs packed with Asian bee pupae and placed them in European hives headed by a European queen, while introducing European pupae to Asian hives, to produce mixed colonies when the adults emerged. Then he tested how the mixed colonies and unmixed colonies responded to waxes produced by both species.

The European bees were not troubled by the Asian bees' wax. They were happy to

extend combs made from either species' wax and the mixed colonies were also able to extend combs made from either Asian or European wax. And when the team analysed the size of the hexagonal comb cells, they found that the European and mixed hives increased the size of cells added to Asian bee comb to the larger European size. However, the Asian bees seemed to be much more sensitive. In hives occupied by Asian bees alone, they would not extend combs made from either wax. The European bees seemed to be more adaptable than the Asian bees and in mixed hives they were also able to get the sensitive Asian bees to build comb under circumstances where they would not have done so if left to their own devices.

But how were the mixed colony workers organising themselves to construct their hybrid combs? Yang gently extracted a honeycomb from a mixed hive and filmed the insects as they worked. According to Yang, natural comb-building honeybees form bridge-like chains that other worker bees stand upon as they apply new wax to cell foundations. Amazingly, when Yang looked at the strings of bees on the combs in mixed hives, he saw bees of both species contributing to the chains and comb construction.

The bees were cooperating regardless of their differences. Despite 2 million years of independent evolution, European and Asian bees still build honeycombs using the same strategy that was originally handed down by their ancestors.

10.1242/jeb.045641

Yang, M.-X., Tan, K., Radloff, S. E., Phiancharoen, M. and Hepburn, H. R. (2010). Comb construction in mixed-species colonies of honeybees, *Apis cerana* and *Apis mellifera*. *J. Exp. Biol.* **213**, 1659-1664.

CHRISTMAS ISLAND RED CRABS REMODEL LEGS FOR LONG HAUL HIKE

No one would be mad enough to run a marathon without any training, but that is essentially what Christmas Island red crabs do every year. They spend months hunkered down, relatively inactive, in inland burrows waiting for the monsoon to come and as soon as the rains hit they're off, marching across the island for up to 12 h a day to the beaches where they mate and reproduce. So how do they pull off this remarkable athletic feat with virtually no training? Having been fascinated by the physiology of these extraordinary crustaceans for much of his career, Steve Morris, from the University of Bristol, knew that the crabs must make major changes to their muscle composition over a very short time to switch from muscle suited to anaerobic short-term sprinting



Ute Postel

(while inland) to aerobic high endurance muscle to sustain their coastal march. Curious to find out what modifications the crabs make, Morris teamed up with Ute Postel, Fiona Thompson, Gary Barker and Mark Viney to find out which genes are expressed most in the legs of crabs before and after embarking on their long haul hike (p. 1740).

Travelling to Christmas Island just before the monsoon broke, Morris and Postel collected leg muscles from crabs while they were migrating and returned 6 months later to collect muscles from the animals during the dry season. Back in the UK, Postel isolated the mRNA (the code molecule that is transcribed from genes and then translated to make protein) from the muscles to see if there were differences between the genes that were transcribed during the monsoon migration and those transcribed when the crabs were inactive during the dry season.

After months building a library from the mRNA molecules from both tissues and analysing the expression levels of many genes, the team could see that there were dramatic differences between the migrating and inactive crabs' muscles. Not surprisingly the majority of the genes expressed in the muscles coded for muscle proteins, such as actin, which forms part of the muscle's contractile unit, and troponin and tropomyosin, which regulate muscular contraction. And, when the team took a closer look at the versions of genes that were expressed, they could see that the immobile dry season crabs' muscles were tuned to short anaerobic sprints while the muscles of crabs that migrated during the monsoon were aerobic, extremely resistant to fatigue and ideal for the crab's arduous odyssey.

The team also found that mRNA transcripts of genes that code for proteins involved in muscle reconstruction were more abundant in the migrating crabs' legs and say 'their comparative up-regulation is consistent with remodelling of leg muscle for migration in the wet season'. So the crabs' muscles undergo a dramatic change as the monsoon sets in, ready for their migration to the coast.

Tragically, Morris did not live to see this work published as he was knocked off his bike on the way into the lab on the morning of 11 August 2009 and died later from his injuries. 'The accident that killed Steve was on the day that the referees' comments came back; he never saw them,' remembers Viney. 'Steve was a traditional physiologist and this was the first time that he had done a study at the molecular level. It was a brave new departure for him and this paper is the first manifestation of this new direction of his research,' says Viney. Reflecting on the impact of Morris's death on his friends and colleagues Viney says, 'Steve never let anything stop him and we took that spirit on to get this paper out'.

10.1242/jeb.045633

Postel, U., Thompson, F., Barker, G., Viney, M. and Morris, S. (2010). Migration-related changes in gene expression in leg muscle of the Christmas Island red crab *Gecarcoidea natalis*: seasonal preparation for long-distance walking. *J. Exp. Biol.* **213**, 1740-1750.

PECKED CHICKENS NOT SINGLED OUT BY ODOUR



Dominic Wright

Chickens have some nasty habits. When cooped up they can begin pecking each other. Poultry farmers would love to know what triggers this antisocial behaviour. Anna-Carin Karlsson and Per Jensen from Linköping University, Sweden, wondered whether the body odour of individual birds may provoke other birds into attacking them, so they approached Matthias Laska, an olfactory physiologist, to find out how realistic this scenario could be. 'Birds are thought to be microsmatic – to have a poorly developed sense of smell,' explains Laska, but when he went digging in the literature he quickly found that some birds rely heavily on their sense of smell. Could Karlsson and Jensen be right? Choosing to test out the body odours of red junglefowl, the common ancestor of all domesticated poultry, the team decided to test whether harassed birds were cursed with a signature scent that singles them out for victimisation (p. 1619).

But first the team would have to find out whether individual poultry have individual body odours. According to Laska, poultry

only have one skin gland, the uropygial – preening – gland, which could produce an individual's odour. Gently collecting droplets of the secretions from the uropygial glands of pecked and unpecked birds Laska decided to test whether mice could distinguish between the odours of individuals.

Training the mice to show that they recognised a smell by taking a drink from a water spout, Karlsson offered a mouse the choice between the odour of the bird that it had been trained to recognise and an unfamiliar odour from another bird. If the chickens smelled the same to the mouse, it would be unable to tell them apart and would respond (by taking a sip of water) to the two odours equally, but if the odours were different, it would respond only to the odour that it recognised. The mice had no difficulties distinguishing between the chickens' odours, so individual chickens can be distinguished based on their odours alone. But could the chickens' odours mark them out as victims?

The team decided to test the mice's responses to pairs of chicken smells. If the mice were trained to recognise a pecked bird's odour (carrying a victim signature), the mice would be able to distinguish between the two odours, because one carried the signature and the other did not, but if the mice were unable to detect the signature they would behave exactly the same as if they were being asked to discriminate between pairs of pecked birds and pairs of unpecked birds. Unfortunately the mice couldn't tell the difference, they were unable to detect a victim signature odour. 'They were too good at distinguishing between individuals,' explains Laska.

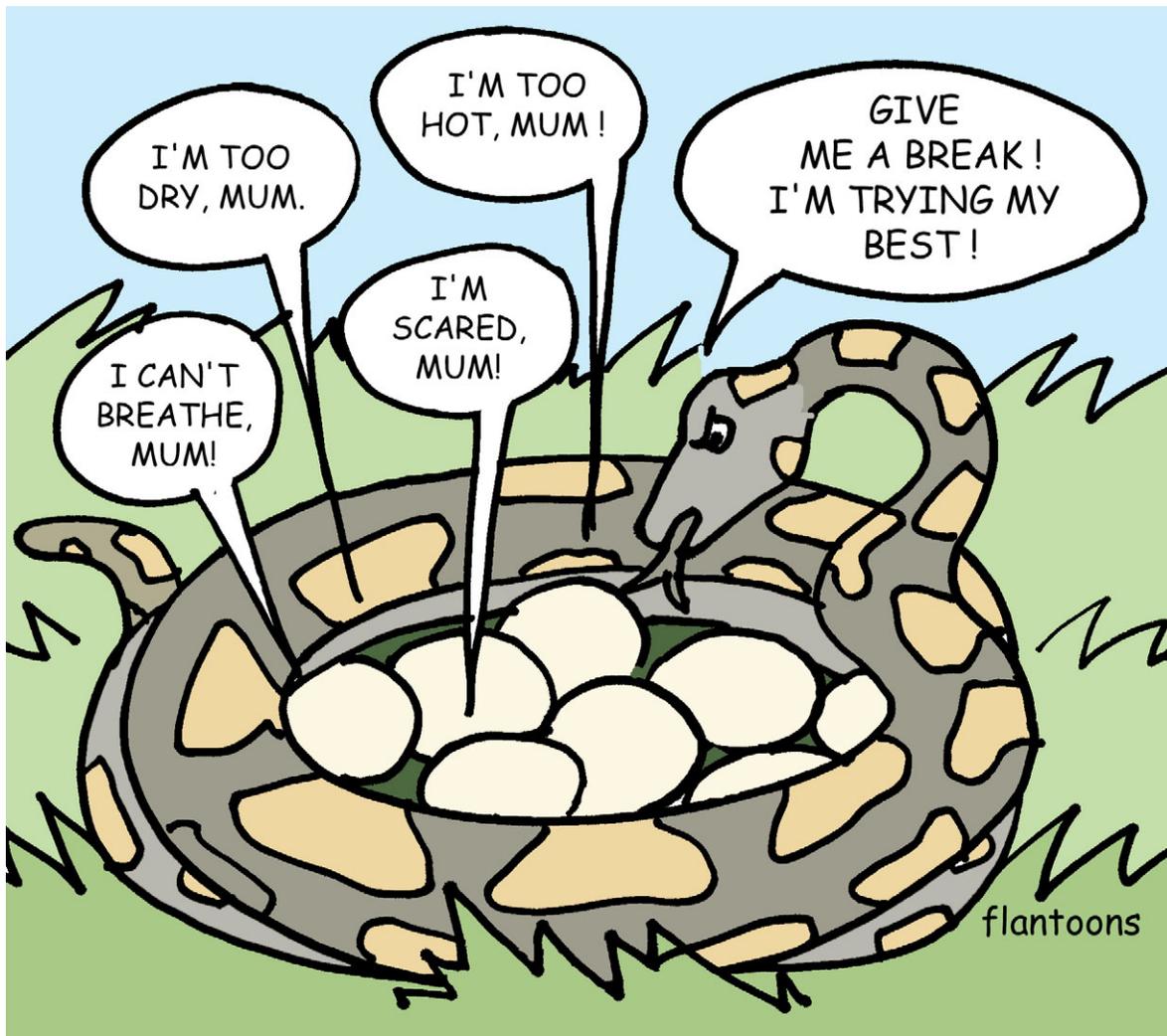
Maybe the mice were missing something, so Laska and his colleagues decided to look at the chemical components of the birds' odours. Teaming up with chemists Mathias Elgland, Katriann Laur, Timmy Fyrner and Peter Konradsson, they analysed the volatile components of pecked and unpecked birds' body odours and found that they were largely composed of fatty acids ranging in length from 10 to 24 carbons. But no matter which way they analysed the fatty acid ratios, there was no signature that could signal a bird out for victimisation.

Although pecked poultry victims are not selected on the basis of their fatty acid smell, it is possible that bullies detect trace materials in pecked birds' odours or even take offence at their feather colour.

10.1242/jeb.045625

Karlsson, A.-C., Jensen, P., Elgland, M., Laur, K., Fyrner, T., Konradsson, P. and Laska, M. (2010). Red junglefowl have individual body odors. *J. Exp. Biol.* **213**, 1619-1624.

PYTHON MUMS HUG EGGS TO PREVENT DEHYDRATION



Some brooding parents take it in turns to care for their clutches, but not pythons. The majority of python mums spend the entire incubation alone, wound around their precious eggs. These mothers protect their clutches from predators and keep the eggs warm in their coils. However, they can also restrict their young's access to oxygen if they clasp too tight, so they periodically relax their grip to improve the eggs' oxygen supply. The developing young are also supplied with all of the water that they will need during development when the egg is laid, but this places the egg at risk of dehydration when conditions are dry. Knowing that incubating python mums adjust their postures to make sure that their young don't suffocate, Zachary Stahlschmidt and Dale DeNardo from Arizona State

University wondered whether they also change position as environmental humidity fluctuates to protect their eggs from dehydration. The duo monitored the behaviour of eight incubating Children's python mums as the temperature cycled between 31.5°C and 26°C at low, medium and high humidities (p. 1691).

Monitoring the snakes' posture and the clutches' temperature and humidity, the duo found that when the air was humid and moist, the mums didn't seem concerned about water loss. They loosened their coils when their eggs were warm and curled them tight when the temperature fell to keep the eggs warm.

However, when the air was dry, the mothers spent significantly more time coiled tightly

around their eggs. Even when the clutch was warm, and the mothers could have relaxed their coils, they kept them tightly wound to protect their young from dehydration.

So python mothers adjust their coiling behaviour in response to humidity and temperature to protect their unhatched young from dehydration and give them the best start in life.

10.1242/jeb.045492

Stahlschmidt, Z. and DeNardo, D. F. (2010). Parental behavior in pythons is responsive to both the hydric and thermal dynamics of the nest. *J. Exp. Biol.* **213**, 1691-1696.

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