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.

CROC JAWS MORE SENSITIVE THAN HUMAN FINGERTIPS

Duncan Leitch

Armoured in elaborate scales, the skins of crocodiles and alligators are much prized by the fashion industry. But sadly, not all skins are from farmed animals. Some are from endangered species and according to Ken Catania from Vanderbilt University, USA, sometimes the only way to distinguish legitimate hides from poached skins is to look at the distribution of thousands of microscopic pigmented bumps that pepper crocodiles' bodies. Adding that the minute dome organs are restricted to the faces of alligators, Catania puzzled, 'What are the organs for?' Explaining that they have been proposed to detect subtle shifts in water salinity and shown to sense ripples in water, Catania says, 'We suspected that there might be more to the story', so he and Duncan Leitch teamed up to take a closer look at the small structures (p. 4217).

Observing the skin of American alligators and Nile crocodiles with scanning electron microscopy, Leitch could see that each dome was surrounded by a hinge depression. And when he sliced through a series of domes to identify the sensory receptor structures beneath, he found sensitive free nerve endings near the dome surface, and laminated corpuscle structures – which are vibration sensitive – and dermal Merkel complexes – which respond to sustained pressure – in the lowest skin layer.

Next, Leitch stained the nerve structures leading from the skin through the reptile's jaw and painstakingly traced the sensitive trigeminal nerve as it branched to the domes. 'The innervation of these jaws was incredible!' exclaims Catania. The entire jaw was infiltrated with a delicate network of nerves. 'There was a tremendous number of nerve endings and each of the nerve endings comes out of a hole in the skull', Leitch adds. Referring to the animal's combative lifestyle, he suggests that this arrangement protects the delicate trigeminal nerve fibres – carried inside the skull – from damage during attacks while maximising the nerve endings' sensitivity at the surface.

But none of these discoveries answered the question of which system the domes relay sensory information to. Recalling that the domes had been proposed to detect salinity changes and even electric fields, Leitch gently bathed the limbs of Nile crocodiles in brackish water while carefully recording the electrical activity in the spinal nerve, but couldn't detect a signal. And when he repeated the experiments while applying a weak electric field to the water, there was no response again. However, when Leitch gently touched one of the sensory domes with a minute hair designed to test human touch sensitivity, he discovered that the domes around the animals' teeth and jaws were even more touch sensitive than human finger-tips. And when he filmed crocodiles and alligators going about their business in the aquarium at night, he was impressed at how fast the animal's 50–100 ms response times were. 'As soon as they feel something touch, they snap at it', recalls Catania.

So, why do such well-armoured animals require such an exquisite sense of touch? Leitch suggests that this sensitivity allows the animals to distinguish rapidly between unpalatable pieces of debris and tasty prey while also allowing mother crocodiles to dextrously aid their hatching young by extracting them from the egg with their jaws. The pair is keen to understand how these sensory areas map onto the forebrain. Explaining that massive regions of the human brain are dedicated to processing touch sensory information, Catania says, 'Crocodilians are not an ancestor to humans, but they are an important branch that allows us to fill in key parts of the evolutionary puzzle for how sensory maps in the forebrain have evolved'.

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Leitch, D. B. and Catania, K. C. (2012). Structure, innervation and response properties of integumentary sensory organs in crocodilians. *J. Exp. Biol.* **215**, 4217-4230.

Kathryn Knight

CUTTLEFISH EMBRYOS LEARN BEFORE HATCHING

Cuttlefish hatchlings are on their own right from the start. Emerging unprotected from the egg, the youngsters have to be able to fend for themselves, capturing food, evading predators and merging with the background if they are to survive. 'The question is, how are they able to perform so many complex behaviours?' says Ludovic Dickel from the Université de Caen Basse-Normandie, France, adding, 'Is this repertoire under genetic control or is the animal able to learn before hatching?' According to Dickel, there is some evidence that the youngsters, which usually prefer to dine on shrimp, can learn to feast


Christelle Jozet-Alves

on crab during the first week of life, but how far back does the ability to learn go? Can tiny cuttlefish embryos that are still developing in the egg learn too? Intrigued, Dickel and his colleagues Sébastien Romagny, Anne-Sophie Darmaillacq, Mathieu Guibé and Cécile Bellanger decided to investigate when the developing embryos' sensory systems begin to function and whether they were capable of learning a simple task (p. 4125).

Collecting eggs on cuttlefish traps, the team waited until the embryos had developed the ability to flex their mantles (stage 23 of development) before testing which of their senses had begun to function. Knowing that cuttlefish hatchlings need to evade European sea bass – they are one of the fish's favourite delicacies – the team tested the embryos' sense of smell by exposing them to the predator's odour, and observed the tiny animals' movements after painstakingly removing the egg's dark outer case. Explaining that the minute cuttlefish pulse their mantles when startled, the team was pleased to see that the developing youngsters flexed their mantles in response to the predators' odour. And when they tested the embryos' sense of touch by gently prodding their mantles with a blunt needle, the tiny cuttlefish also reacted. Even at this early stage of development, the embryos had developed the senses of smell and touch.

Yet, when Dickel and his colleagues tested the embryos' reaction to light, the animals barely stirred: their visual sense had not developed sufficiently. However, when the team repeated the test 1 to 2 weeks later when the visual pigments had developed (stage 25 of development), the cuttlefish pulsed their mantles: the visual system was finally functioning. 'The visual system is also the last system to mature in vertebrate embryos, so this is an impressive homology between vertebrates and cephalopods as they diverged very early during evolution', says Dickel.

Having pinpointed when the tiny cuttlefish begin to perceive light, the team tested whether the animals could learn to become desensitised to light. As soon as the embryos' visual system was functional, the team showed the youngsters 150 s long flashes of light interspersed by 30 min intervals of darkness: the youngsters enthusiastically pulsed their mantles whenever the light came on; they were unable to learn to ignore it. However, by stage 30, the embryos picked the idea up quickly. They soon began to lose interest in the light and when the team checked that the older embryos simply weren't tiring of the task – by throwing in a gentle tap from the needle between the bursts of light – the animals regained some of their interest in the light and began pulsing more energetically again. The cuttlefish embryos were not tiring, they were learning. So, cuttlefish embryos are able to collect information while in the egg, and Dickel is keen to find out how much sensory detail the tiny embryos can process before they embark on life in the open.

10.1242/jeb.081943

Romagny, S., Darmaillacq, A.-S., Guibé, M., Bellanger, C. and Dickel, L. (2012). Feel, smell and see in an egg: emergence of perception and learning in an immature invertebrate, the cuttlefish embryo. *J. Exp. Biol.* **215**, 4125-4130.

Kathryn Knight

CRABS SMELL FEAR THROUGH ANTENNULES

Even if you're armed with a fierce pair of pincers, life can be risky. There's always going to be a larger crab that might decide to dine on you. So, crabs tend to give the odour produced by injured crabs a wide berth, treating it as a warning that a larger predator may be lurking nearby. 'The environment is filled with threats,' says Marc Weissburg from the Georgia Institute of Technology, USA, although it is also full of the scents of enticing tasty treats. 'Unless you are immune to eating, then you are always going to have to confront the conflict between aversive and attractive odours', Weissburg says. Intrigued by how crabs successfully negotiate this conflict, Weissburg and a team of undergraduate student researchers set out to discover which course blue crabs (*Callinectes sapidus*) steer when presented with a whiff of lunch spiced with danger (p. 4175).

Collecting crabs from the ocean off Savannah, Georgia, Weissburg returned with them to the lab where he and Kimberly Berkenkamp started to test their reactions to plumes of different odours in a flow tank. Sure enough, when the animals were downstream of luscious shrimp aroma they practically galloped toward the origin. However, when the water smelled of injured crab, the animals became evasive and some even buried themselves to avoid the stench.

But, how would they react when the two odours were flowing in parallel in close proximity? This time, the crabs were much more cautious. Although they continued to pursue the attractive scent, the crabs actively avoided the side of the tunnel that smelled of injured crab. Discriminating between the plumes and following the attractive odour to its source, the crabs were able to successfully home in on the food despite the close proximity of the warning signal.

However, when the duo disrupted the flow by introducing a cylinder into the water, to generate turbulence and mix the plumes, Weissburg says, 'The animals were no longer willing to track to the attractive source and they reacted as if there was only an aversive source'. Thoroughly mixing the two odours had altered the crab's perception of the tasty prawn aroma sufficiently for it to no longer appear attractive. Weissburg decided to find out which of their many olfactory organs the crabs use to distinguish between the attractive and aversive odours.

'In blue crabs, the control over orientation toward an attractive substance is split between the antennules, which are the small structures between the eyes on the head, and chemosensors on the tips of the walking legs', explains Weissburg. So, together with Lauren Atkins and Danielle Mankin, he tested the animals' reactions to the merged plume when one or other of the 'nostrils' were effectively plugged.

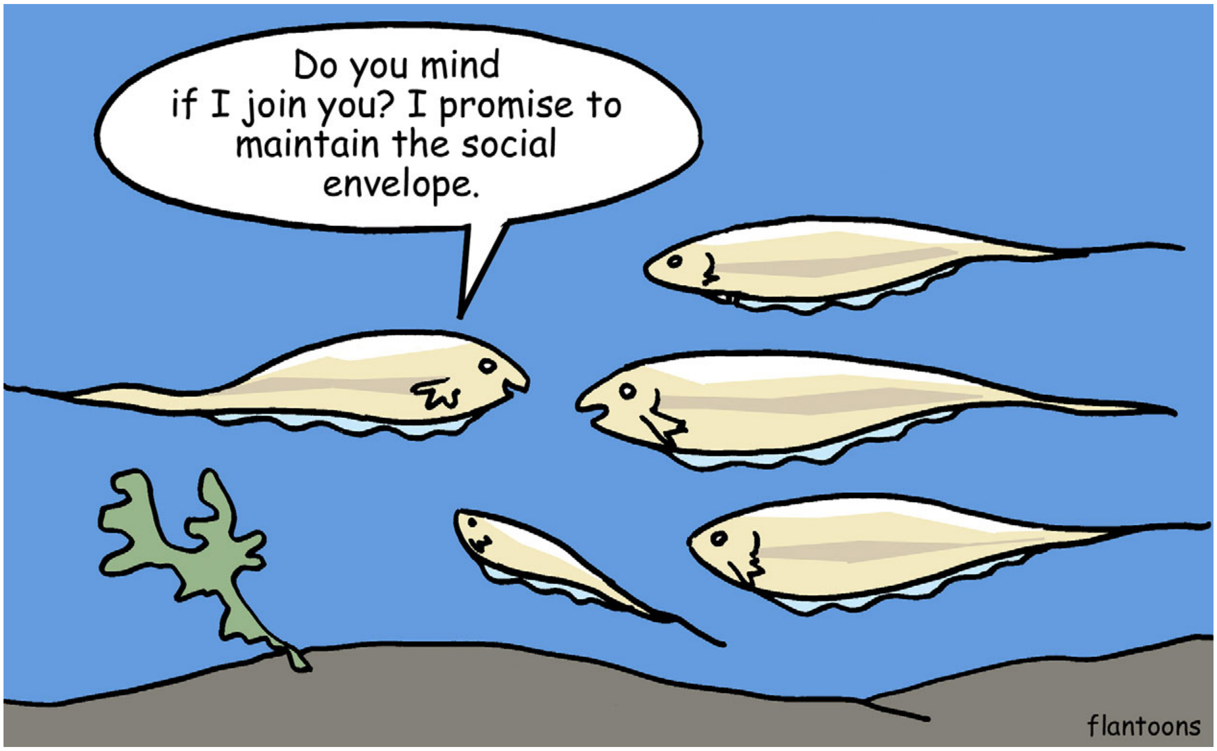
Desensitizing the scent receptors on a crab's legs by immersing them in fresh water, Weissburg then released the animal into the blended plume: the animal tried to take cover. However, after gently removing the scent receptors from another crabs' antennules and positioning it in the plume mixture, the animal bounded toward the source of the attractive aroma of food as if it was uncontaminated. The crab was no longer repelled by the aversive odour. 'We had knocked out the input [from the antennules] that suppresses navigation. The behaviour of the animals to track the attractive cue in the presence of the aversive cue was rescued', explains Weissburg.

10.1242/jeb.081968

Weissburg, M., Atkins, L., Berkenkamp, K. and Mankin, D. (2012). Dine or dash? Turbulence inhibits blue crab navigation in attractive–aversive odor plumes by altering signal structure encoded by the olfactory pathway. *J. Exp. Biol.* **215**, 4175-4182.

Kathryn Knight

GREGARIOUS ELECTRIC FISH ADJUST TO MAINTAIN SOCIAL ENVELOPE



Bathed in their own gently oscillating weak electric field, glass knifefish (*Eigenmannia virescens*) interpret distortions of the field to learn about their surroundings, although the fish rarely enjoy the opportunity to analyse perturbations in their isolated fields. According to Sarah Stamper and Manu Madhav from Johns Hopkins University, USA, these fish spend most of their time in groups and in order to avoid jamming each other's electric fields with their own oscillations, the fish adjust the frequencies of their fields to prevent a clash. In addition, the team explains that the complex oscillating field that results when the fish congregate is enfolded by an envelope – known as the social envelope – which also has a characteristic low frequency ripple. Curious to find out whether glass knifefish are able to identify the social envelope produced by a small crowd and modulate their own electrical fields in response, Stamper, Madhav, Noah Cowan and Eric Fortune simulated the electric fields produced by two glass knifefish and measured the response of a third fish as they systematically varied the field's social envelope (p. 4196).

Presenting a fish with a simulated composite field with a low frequency (2 Hz) social envelope, the team found that the fish altered the frequency of its own oscillating electric field significantly, although fish located in a field with a higher frequency social envelope (4–8 Hz) responded more weakly. Thus, the fish were able to detect low frequency social envelopes. And when the team analysed the impact of the third fish on the structure of the social envelope, they realised that the additional fish raised the frequency of the social envelope to 5–15 Hz. However, the fish were not simply increasing the frequency of the oscillating field to avoid jamming the signals of the other nearby fish. The team suspects instead that the fish increase the social envelope frequency to improve their electrical perception, as very low frequency electric field envelopes (~2 Hz) may impair their ability to perceive their surroundings and objects moving in the vicinity.

10.1242/jeb.081976

Stamper, S. A., Madhav, M. S., Cowan, N. J. and Fortune, E. S. (2012). Beyond the jamming avoidance response: weakly electric fish respond to the envelope of social electrosensory signals. *J. Exp. Biol.* **215**, 4196-4207.

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