OUTSIDE JEB

Dolphins show their age differently

'What? You're 40? You don't look a day over 30!' This is sweet music to some of our ears! We all know that every animal is born, grows, matures and ages in a predictable way. After all, ageing is part of the life cycle of every living thing on Earth. Yet, scientists still do not have a clear picture as to why some people do not look their age, while others age much faster than the majority. Is age just a number or is it much more than that?

To answer this question, veterinary epidemiologist Stephanie Venn-Watson from Epitracker, Inc., CA, USA, along with a team of researchers from institutions in California and Arizona, USA, turned to studying ageing in captive bottlenose dolphins. In addition to being extremely lovable, dolphins are a great study organism because they are long lived and show similar afflictions to humans when they age, such as loss of memory, inflammation in the joints, anaemia (or low blood iron levels) and type II diabetes. Working with the US Navy Marine Mammal Program – an organization training and caring for aquatic mammals – the research team accessed blood samples collected from dolphins from 1994 to 2018. The group was particularly interested in how blood composition changes over time and whether that correlated with ageing rates in 44 dolphins.

The scientists found that haemoglobin, which is an iron-containing protein found in red blood cells that carries oxygen to tissues, declined throughout the lifespan of the dolphins, leading to low blood iron levels and anaemia in old age. In fact, the group concluded that faster ageing dolphins, meaning those that showed more markers of ageing than others, had larger declines in blood haemoglobin than those dolphins that aged slower over a period of 30 years. Anaemia is also a characteristic of ageing in humans, often referred to as 'anaemia of old age'. The work conducted by Venn-Watson and her colleagues suggests that not only are dolphins a great model to study human ageing but also that the ageing rate itself may depend on declines in blood iron levels over time.

In addition, the researchers found that platelets – small cells in the blood that help form clots – along with lymphocytes, which are white blood cells involved in the body's immune response, can also be used as markers of ageing rate in bottlenose dolphins. Both platelet and lymphocyte levels declined in fast ageing animals, while in those with slower ageing rates, the decline over 30 years was much slower. Interestingly, the authors did not find any differences in ageing rates between male and female dolphins, even though the females are known to live longer in the wild. The authors attributed this to the lifestyle of the animals: here, they were kept in captivity and were well cared for, while in the wild, the males are more exposed to predation because of their vast territory, and are more aggressive than the females and, therefore, more prone to injury by other males, making them more at risk of dying.

In their study, Venn-Watson and her colleagues found that bottlenose dolphins, much like humans, age at different rates, despite being held in identical conditions. Those animals with faster ageing rates also tended to be more susceptible to age-related conditions, such as anaemia and a compromised immune system. These findings may dare scientists to look at the genetic code behind these ageing systems, to try to slow down ageing in humans or, at the very least, improve the quality of life in the ageing population. It may be the case that in the not too distant future, age will, indeed, be just a number.

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When life gives you food, grow more arms

The body plan for most animals is set at birth. For humans, we get two arms, two legs and no more. It doesn't matter if you lose one, or if you really want another hand to hold your coffee, our genetic code decides how many appendages we have. But research led by Aissam Ikmi from the European Molecular Biology Unit in Germany has recently shown that sea anemones can sprout extra limbs when they are given lots of food. The team, which also included researchers from the University of Kansas and Stowers Institute for Medical Research, USA, wanted to explore how these squishy little animals can be so flexible with their body plans and what molecular machinery was causing these huge changes.

Ikmi's team focused on the starlet sea anemone (*Nematostella vectensis*), fascinating animals because they can branch out to grow new body parts across their lives – kind of like plants. Starlet sea anemones have quite simple body plans, with a bulbous, oblong body and some squirmy tentacles (or arms) at one end that help them capture prey. Researchers have long admired the flexible lives that these

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little creatures lead, but it was unclear what triggered the anemones to sprout more arms. Ikmi suspected that food might fuel this growth. To test the idea, the researchers fed some anemones daily brine-shrimp snacks while others were left empty handed. The fed anemones all started out with four arms, doubled their size and then budded new pairs of arms after about 5 days, some eventually grew up to 16 arms.

But, how does a meal get turned into new arms? The research group looked at two key components of the mechanism that manages growth to answer this. They first measured the target of rapamycin protein (TOR), because it regulates cell growth based on the level of nutrients and energy available. Only the fed anemones had a lot of TOR activation and the TOR activation shifted to places where new arms would soon pop out with each passing day of brine-shrimp munching.

The team next investigated fibroblast growth factor receptor (FGFR) as a potential second cog in the arm-growing machine and they found the gene for this receptor was only expressed where a new arm would unfurl. When the researchers chemically blocked the receptor or rendered the gene functionless, the anemones would still grow in size, but they couldn't sprout any new arms. This suggested that feeding tells TOR to first make the anemones grow, which then triggers FGFR to be expressed at new arm sites. TOR activity then becomes more localized, and voilà, new arms shoot out.

Ikmi analysed over 1000 sea anemones to show that TOR and FGFR must volley back and forth to transform a feeding frenzy into new appendages. Their elegant series of experiments traverses many scales of biological organization, from genes to environmental cues, and these sea anemones certainly give a whole new meaning to the consequences of over-eating.

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Prolactin makes rodent dads care for their rug rats

Most mammals relegate all parental duties to mom. However, some mammal dads, such as common house mice (*Mus musculus*), do share caregiving duties. Male mice normally bully unrelated babies, but soon after they sire their own offspring, they pitch in with caretaking. In contrast to mice, rats (*Rattus norvegicus*) never develop a parenting instinct, even after having a litter of their own. Intrigued by this disconnect in fatherly involvement between murines, Stefanos Stagkourakis from Christian Broberger's lab at the Karolinska Institutet, Sweden, in collaboration with an international team of colleagues from Austria and New Zealand, explored why these rats and mice take on such different dad duties.

The hormone prolactin is well known for spurring milk production and parental care in mammalian moms, but its role in paternal behavior was largely unknown. Stagkourakis wondered whether prolactin might explain why male rats, but not male mice, neglect their young. Knowing that dopamine prevents prolactin release, Stagkourakis measured how often male rats and mice release this neurotransmitter. They found that the rat neurons released more dopamine, resulting in dramatically lower prolactin levels in the blood compared with male mice. These results suggest that rats' higher dopamine and consequently lower prolactin levels make them unmotivated fathers. As such, perhaps all that rat dads need to make them as fatherly as mice is more prolactin.

Based on these findings, Stagkourakis and his team next asked a fairly straightforward question: does boosting rats' prolactin levels to rival that of male mice make rats better dads? To test this, Stagkourakis placed virgin or recent rat dads in an arena with several pups and gave some of the dads a shot of prolactin to see how attentive they were towards the needy pups while parenting solo. Compared with rat dads that didn't get a prolactin boost, rats that did spent more time grooming pups and huddling over them. Importantly, the virgin males were unaffected by prolactin injections, which suggests that sexual experience is required to make male rats receptive to prolactin's parental influence. In other words, prolactin can quickly induce parental behavior; however, it was unclear whether preventing prolactin's action would cause fatherly mice to relapse into deadbeat dads.

For their final set of experiments, Stagkourakis and colleagues determined whether prolactin inspires parental involvement by mouse fathers. To do this, the team removed the prolactin receptors from the hypothalamus of some mouse dads and checked the animals' parenting skills. Instead of their usually high involvement, these dads became less fatherly, spending less time attending to the pups. In addition, Stagkourakis used a technique known as optogenetics to activate dopamine neurons precisely in the brains of a few male mice to mimic the rat's pattern of dopamine release, triggering rat-like prolactin release, and again the mice reduced the care they gave their pups. So, rodent dads rely on prolactin to activate parental care-inducing brain cells in the hypothalamus.

Overall, Stagkourakis and colleagues have found that rats and mice are both equipped with the tools to be either deadbeat dads or fathers of the year. However, it turns out that it's prolactin that makes the parent.

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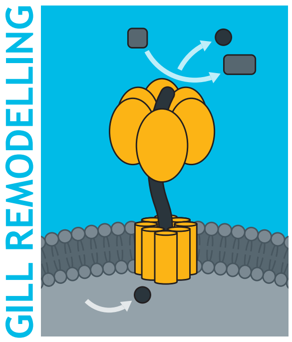
Blood-thirsty lamprey transform their gills



Sea lampreys are masters of transformation. These ancient fish begin life as tiny worm-like creatures that bury in sandy riverbeds and filter food particles out of the water. However, after years of sedentary existence, lampreys undergo a dramatic metamorphosis; just like caterpillars turn into butterflies, lamprey larvae turn into, well, something less gorgeous. The fish grow larger, become active eel-like swimmers and leave their sandy homes to migrate out to sea. Once in the ocean, filter-feeding is passé and lampreys transform their mouths into a suction cup with alien-looking, circular rows of teeth that latch on to larger fish, pierce their skin and suck their blood; a rare case of parasitism in a vertebrate. However, these lifestyle changes come at a price. The saltier water in the ocean requires a complete overhaul of the gills, which handle the internal salt balance of fish. And, the digestion of a blood meal, which is extremely rich in protein, produces toxic ammonia that needs to be cleared into the water, through the gills. Julia Sunga and her colleagues at Wilfrid Laurier University, Canada, set about investigating how lampreys remodel their gills during metamorphosis to accommodate their new parasitic lifestyle.

In the lab, the team either held maturing lampreys in freshwater or transferred them to seawater to simulate their natural ocean migration. In addition, to study the change in diet, the team kept one group of lampreys in a tank together with some hapless rainbow trout that would serve as prey. The researchers then measured how much toxic ammonia had built up in the lampreys' blood and how much they released into the water.

Over the course of the lampreys' development, the quantity of toxic ammonia in their blood increased and was highest after metamorphosis. However, the amount of ammonia released into the water changed little throughout metamorphosis and was unaffected by whether the animals were held in freshwater or seawater. Even more surprisingly, those parasitic lampreys that were feasting on the blood of trout released 5 times more ammonia into the water than the lampreys that were not dining and this kept the levels of ammonia in their blood very low. To find out how, the researchers zoomed in on their gills.

Immunohistochemistry revealed that larval lamprey gills were covered in rhesus glycoproteins, small channels that can funnel ammonia across cell membranes. The researchers also found a proton pump that generates acidic conditions around the channels to promote the removal of ammonia from the lampreys' blood, expelling it into freshwater. However, during metamorphosis, these proteins began to disappear. Instead, new cells started to develop, packed with power-generating mitochondria, which would maintain the lampreys' salt balance in seawater. These new cells had their own ammonia channels, only this time, they were linked to a sodium/potassium pump. Under the microscope, the two proteins seemed to overlap perfectly within the cells, especially in lamprey that had been feasting on blood. Thus, it seems that lamprey have a broad toolkit at their disposal to clear ammonia from their blood, which they adjust throughout development, based on the salinity of the water and their preferred diet.

Lampreys are ancient creatures that pre-date the evolution of jaws and their intricate life cycle spans a broad range of habitats, appearances, behaviours and diets. This close look at the gills of developing lampreys revealed the surprisingly flexible machinery that allows the versatile creatures to maintain salt balance and ammonia excretion throughout their changing lifestyle. A lamprey's transformation may be less pretty than that of a butterfly but, nonetheless, parasitic lamprey are awe-inspiring creatures, just in a nightmarish kind of way.

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Fish at the spa: too relaxed to care

If there was a monstrous night-time thunderstorm and the power went out in your home, all during a zombie apocalypse, you would probably freak out at the slightest unusual noise and flee to your panic room. However, if you'd just got back from a relaxing hot stone massage at the spa, had cozied up in a fuzzy bathrobe with a chilled glass of buttery chardonnay and were watching a soothing nature documentary when you heard an unusual noise, you would probably ignore it and attribute it to pipes, the wind or something benign. Your condition can affect how you perceive threats, but is it the same for fish? To figure this out, a team of scientists led by Vinicius Giglio of the Federal University of São Paulo, Brazil, devised a cleverly simple experiment.

Fish do not drink wine to relax and they cannot wear fuzzy bathrobes, but they do go to spas – of sorts. At many coral reefs, there are cleaning stations where little fish – often teeny tiny wrasses with a black stripe – pick parasites and dead skin from the skin of larger fish, sea turtles, or whoever wants to be cleaned. The client gets to feel clean, healthy and smooth while the cleaner gets a delicious snack. Everyone wins, or at least seems to win. But is there a negative tradeoff for the client?

To test this, Giglio had snorkelers go to coral reefs in a marine reserve off the coast of Brazil to chase parrotfish and squirrelfish – some of which were out and about, while others were getting pampered at cleaning stations. The snorkelers tried to see how close they could get to the fish before the animals swam away, recording that distance as the ‘flight initiation distance’. Then, the team compared the behaviors of the two groups to see whether spa treatments affect fish behavior.

It turned out that the fish at cleaning stations would allow snorkelers to approach closer than those just doing average fish stuff. While respectful snorkelers are not likely a serious threat to reef fish, they do simulate predators. Therefore, reef fish at cleaning stations may allow Bruce the shark – of Nemo fame – to get too close for comfort, but why might these fish lower their guard?

Giglio has three potential answers. One is that fish at cleaning stations need to weigh the benefits of being picked clean of parasites against the danger of being eaten; leaving the spa early means missing out on the full treatment, so fish may take a risk and allow predators to get closer before fleeing to get the most out of their cleanup. Another reason could be that cleaning stations may be governed by a peace treaty, which allows them to act as safe havens where fish do not eat other fish, because aggressive behaviors would ruin the spa for everyone, including predatory fish that also wish to use them. The last reason may be that the gentle grooming of cleaners is soothing to reef fish and may lower levels of the stress hormone cortisol; because these fish are less stressed, they may be less likely to perceive a threat as such.

Regardless of the reason, client fish at cleaning stations perceive threats differently. However, feeling safe and being safe are two very different things. Therefore, getting groomed at a fish spa may be a double-edged sword that leaves the client more vulnerable to being eaten. Next time you are relaxing, make sure not to let your guard down, or else Giglio’s snorkelers may get the drop on you!

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