OUTSIDE JEB

Hey bumble bee, did you have a good year?

Environments are everchanging, forcing animals to deal with daily and seasonal variations, such as changing temperature or rainfall. Such variations can shift the times when different species are active and can affect what foods are available, causing direct and indirect competition with other animals for limited resources. Environmental shifts can have different effects on an animal depending on its life stage. Yet, these important life-stage-specific responses to environmental change are often unaccounted for in ecological studies, not because scientists do not recognize their importance, but because long-term monitoring can be challenging. However, these types of studies are desperately needed if we are going to predict the vulnerability of a given species to climate change. Jane Ogilvie at the Rocky Mountain Biological Station, USA, and Paul CaraDonna at the Chicago Botanic Garden, USA, asked how bumble bees at each life stage respond to changes in weather, pollen and nectar availability and how the abundance of previous life stages affect the following generations.

From 2015 to 2021, Ogilvie and CaraDonna monitored a community of seven bumble bee species every week between April and September in the Colorado Rocky Mountains, USA. They recorded the numbers of worker bees, emerging queens and males, while noting whether the insects were foraging, just flying through, looking for new nesting sites, or whether the males were searching for unmated females. The pair also recorded the temperature and precipitation at each site, to find out how the weather conditions influenced the activity of workers and male bees. As queens can only come out of hibernation once the ground is completely free of snow, Ogilvie and CaraDonna recorded the date when the snow melted completely, to look for connections between how long each winter lasted and queen bee winter survival. Finally, because bumble bee species visit various different flowers, the pair tallied and identified the flowers at each site and counted how many times workers of each species visited each kind of flower to see how changes in pollen and nectar availability influenced bumble bee population sizes.

Ogilvie and CaraDonna discovered that bumble bee species respond differently to changes in weather and food availability. For example, longer winters reduced the number of queens that survived to spring, presumably because they burned through their winter energy stores, starving to death. In addition, only three species in their study – *Bombus flavifrons*, *Bombus insularis* and *Bombus mixtus* – showed an increase in the number of males flying during warmer, drier weather. Yet, the workers of all species but one – the nest parasite *B. insularis*, which doesn't produce workers of its own – seemed unaffected by the weather. They also found that worker-bee numbers increased when there was more pollen and nectar available and, with more workers collecting pollen and nectar, the colony had enough food to create more queens and males to venture off and establish new colonies. And, for some species, such as *B. mixtus*, the number of bumble bee workers from the year before determined how well the overwintered queens did the following year.

This investigation by Ogilvie and CaraDonna has implications far beyond studies of social insect pollinators. The study emphasizes the need for researchers to account for life-stage-specific sensitivity to shifts in weather conditions and food availability if we are to have any hope of predicting how a given animal will fare in a changing world.

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Ogilvie, J. E. and CaraDonna, P. J. (2022). The shifting importance of abiotic and biotic factors across the life cycles of wild pollinators. *J. Anim. Ecol.* **91**, 2412-2423. doi:10.1111/1365-2656.13825

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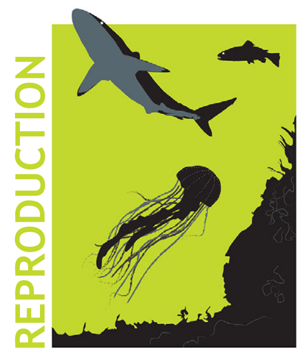
Ricefish plugs keep eggs snug



Fishes care for their young in a variety of ways. While some fishes are 'transfer brooders', which forget about their young after laying their eggs, others are 'pelvic brooders' and carry their eggs for long periods of time, allowing the embryos to develop and grow while protected by their parent. Ricefish have evolved a peculiar sticky structure that emerges from the genital pore and holds their eggs on the outside of the body. This 'plug' keeps eggs safe until hatching then deteriorates so that the fish can prepare for its next spawn. However, we know little about the anatomical changes in the reproductive system of fishes that have allowed some to carry their eggs, in contrast to those that just lay their eggs, or how the plug evolved in the first place. To understand how the plug forms, Alina Schüller from Museum Koenig, Germany, and an international team of colleagues documented the anatomy of a *Oryzias eversi*, an egg-carrying ricefish during the spawning period. They aimed to

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categorize which cell types are involved in plug formation and how the anatomy of these fish changes across time.

The researchers collected *O. eversi* females at various points in the spawning process and examined their reproductive tract and egg bundles attached to the plug using a microscope. They also CT scanned the abdomens of the ricefish on two occasions: in the late stages of the spawning cycle and after the eggs had hatched and left the mother. They compared the anatomy of these egg-carrying species with a close relative, *Oryzias latipes*, which doesn't carry its eggs.

The team discovered that the plug-like structure is made up of filaments anchored within the tube that delivers the eggs from the ovary to the outside world and the reproductive anatomy of the two ricefish species didn't differ much between the egg carriers and those that simply lay their eggs. However, the entire spawning process is much slower for egg-carrying species. While the egg-laying species can develop new eggs within the ovaries while spawning, the egg-carrying species cannot simultaneously form new eggs while still carrying the previous clutch. The plug also prevents the ovary from becoming infected by closing the tube that leads from the ovary to the outside world. Interestingly, the researchers found giant cells with multiple nuclei within the plug used to anchor the eggs of their mother. Even more intriguingly, humans also produce these specialised cells, called macrophages, when we pick up a splinter, or some other foreign body, and our skin swells and reddens as part of an inflammatory response, suggesting that formation of the plug may have evolved from an immune response.

Learning how some mechanisms in the body, such as immune responses, can change in their function for new uses, such as new reproductive strategies, can help us to better understand the origins and intricacies of these mechanisms. This knowledge can help scientists to establish the relationships between various species and how diverse animals such as fishes are, not only in their anatomy, but also in the ways that they live and reproduce.

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Schüller, A., Vehof, J., Hilgers, L., Spanke, T., Wipfler, B., Wowor, D., Mokodongan, D. F., Wantania, L. L., Herder, F., Parenti, L. R., wamatsu, T. and Schwarzer, J. (2022). How to stay attached – Formation of the ricefish plug and changes of internal reproductive structures in the pelvic brooding ricefish, *Oryzias eversi* Herder et al. (2012) (Beloniformes: Adrianichthyidae). *J. Morphol.* **283**, 1451-1463. doi:10.1002/jmor.21518

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Fertilizers disrupt the electric feel of flowers

Electricity is literally all around us. As humans, we generate weak electric fields whenever we move our muscles; spiders use static electricity in the air to fly across great distances; and geckos use static in their sticky feet to scale vertical walls with ease. Since everything has an electric field, we can imagine that this fundamental force of nature plays an important role in many ecological interactions. Among insects, bumblebees are known to alter the static electricity of flowers to communicate with members of their hive; the positive charge that collects on their tiny hairs alters the electric field around a visited flower for ∼100 s, communicating to the next bee that buzzes by that the nectar and pollen rewards from that flower have already been exhausted. Ellard Hunting and colleagues from University of Bristol, UK, and the Woods Hole Oceanographic Institution, USA, set out to test whether the application of agricultural sprays alters the electric field of flowers and whether bumblebees can detect these disruptions.

The pervasive use of chemicals in agriculture is a substantial source of pollution. But besides chemical toxicity, little is known about how agrochemicals otherwise affect plant–pollinator interactions. Hunting and colleagues observed that bumblebees tend to hover around chemically treated flowers and decide not to land. To investigate the cause of this avoidance behaviour, the researchers developed a computational model for how the application of agricultural sprays may alter floral electric fields up to 4 cm from the flower. Their model considered how the presence of droplets on the flower's surface and localized increases in humidity from a spray application might impact the flower's electric field.

Next, the researchers studied the effect of common agricultural fertilizers on flower features that bees recognize. To see if fertilizer sprays change the way bees see flowers, the researchers determined what wavelengths of light are reflected from droplets with and without fertilizers added. To study whether fertilizer sprays change the way flowers smell, they measured how much effort the bees put into feeding when offered sugar water laced with fertilizer compared with just sugar water. Lastly, to visualize the electric fields surrounding various blooms, the scientists sprayed charged coloured powder particles onto the centre of ragwort flowers and observed how the powder distributed across the surfaces. Although the fertilizers did not affect how bees see or smell flowers, the chemicals did alter the electric field around each bloom for several minutes.

Next, Hunting and colleagues tested whether the adverse effects of spray applications persist over time. To do this, they planted electrodes into the cut stems of potted lavender and Texas bluebell plants and recorded the electrical current streaming down the stems in response to different spray applications. While a water spray caused a change in current that only lasted 30–60 s, a spray containing the fertilizer caused a consistent change in current that lasted up to 16 min. A spray with another common agricultural chemical – the pesticide Imidacloprid – induced a change in current that lasted up to 25 mins. Even subsequent water sprays, simulating rain, produced prolonged changes in the plant stem's electrical signature, suggesting that this effect can endure even after a single application of fertilizer.

Finally, the researchers observed wild bumblebees foraging on cut flowers with artificially applied electric fields to mimic the effect of fertilizers. They pierced lavender stems with metal electrodes, placed them in glass jars of tap water, and attached a 13 V battery to produce an electric current in the plant. By recording 2 h videos and counting the number of bumblebees that approached versus those that landed on the lavender, the team showed that bumblebees somehow sense weird electricity as they approach a flower and land less as a result, serving as a healthy reminder that as humans we affect our environments in ways we can't even sense.

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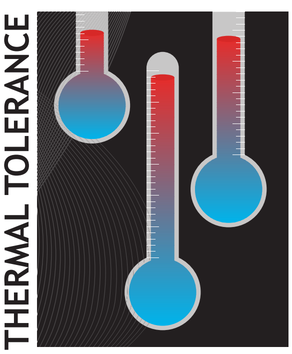
Does turning up the heat affect how salmon eat?



Chinook salmon have tremendous cultural, economic and ecological value in North America – plus, they're delicious. Unfortunately, at their southern range limit in central California, Chinook face several challenges that threaten their survival. One of the most concerning is the large swings in temperature they currently experience, which are expected to worsen due to climate change and human alterations to river water flow. The good news is that managers can help the fish by strategically releasing water from dams to control the temperature of their freshwater habitat. In order to know when, where and how much water to release into the rivers, managers need to know what temperatures keep the fish happy and healthy. Researcher Vanessa Lo at the University of California, Davis, USA, worked with colleagues from the University of California, Davis, the University of Amsterdam, The Netherlands, and NOAA Southwest Fisheries Science Center, USA, to inform Chinook conservation efforts in California by determining which temperatures the fish thrive in.

To accomplish this, Lo and the team measured how temperature impacts the amount of energy the Chinook need to survive and use for their digestive processes. Just like it takes money to make money, it takes energy to break down food into nutrients and fuel. The cost of digesting a meal takes up a substantial portion of a fish's energy budget. If temperature causes these digestive costs to increase, fish may have less energy available to swim and grow. This is especially concerning for young Chinook, which must grow up in their cool freshwater rearing grounds before migrating out to sea through warmer water. If the young fish are unable to make it to the ocean, the population could be lost. So, Lo and colleagues set out to find the temperatures where young Chinook's digestion was impaired to help managers set fish-friendly river temperatures. The team anticipated that as the temperature increased, the time it would take for the fish to digest a meal would decrease. At the same time, they expected that the total cost of digesting a meal would remain the same across temperatures. However, they also predicted that the fish would have to use up a larger proportion of their energy budgets while digesting a meal in warm water, to fit the same overall digestive costs into a shorter amount of time.

The researchers fed young Chinook a meal of fish pellets and then measured how much oxygen the fish were breathing to estimate their energy costs during digestion. They then repeated the procedure at temperatures ranging from 13 to 24°C and, surprisingly, their findings were the opposite of what they had expected. Instead, the time it took for the Chinook to digest a meal and their energy usage during digestion was exactly the same across the entire temperature range, up to 24°C. These results indicate that temperatures up to 24°C do not limit the young Chinook's ability to eat, swim and grow. However, when the team raised the temperature to 25°C, the fish began to struggle so much that they called off the tests.

It seems that juvenile southern Chinook are well positioned to deal with a wide range of temperatures; however, they hit a physiological wall near 25°C. And the longer a fish is exposed to a stressful temperature, the more problems it will develop. Given that the team performed all their energy use measurements after each animal had been briefly exposed to one of the temperatures, they point out that longer exposures to temperatures below 25°C could also negatively affect young Chinook. However, in the short term, Chinook could consume a meal at temperatures beneath 25°C and still have plenty of energy left over for all the other fishy activities they need to survive.

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