

INSIDE JEB

Veggie fish depend on bacterial detox backup but carnies do not



A rainbow darter (*Etheostoma caeruleum*). Photo credit: Jeff Finley, USFWS (public domain).

Protein in the diet is essential for building muscles, skin, cartilage and bone, but there is a drawback to this key dietary component. 'Protein catabolism during digestion generates an appreciable level of ammonia in the gastrointestinal tract', says Leah Turner from York University, Canada. The problem is that ammonia is extremely toxic for fish, so they excrete 85% of the poisonous gas for safety, while converting the remaining 15% into other less toxic components for conversion into proteins. Knowing that many key digestive enzymes are created within the intestines, PI Carol Bucking, also from York University, suspected that this might also be the case for ammonia detoxification enzymes, but she wasn't entirely sure that it was the whole story. 'They [ammonia detoxifying enzymes] may also be generated by the bacteria that inhabit the gastrointestinal tract', she says. In addition, she knew that carnivorous species are more likely to be vulnerable to ammonia poisoning than vegetarian species, thanks to their protein-rich diet. Intrigued by the possibility that a fish's gut flora may help out, the duo decided to test how one

carnivorous species (the rainbow darter – *Etheostoma caeruleum*) and a veggie species (the central stoneroller – *Campostoma anomalum*) detox the ammonia generated by their diets.

'Catching stonerollers was easy, but we had to catch the darters by hand. It took a while to hone our technique', chuckles Turner, who worked with Bucking to select fish from three different locations on two rivers – Irvine Creek and Lutteral Creek in Ontario, Canada, 'We wanted to choose areas that were geographically close but had different features agricultural land versus forest - that could be differentially affecting the microbiome [bacterial colonies in the intestine]', says Turner. Back in the lab, she analysed the activity of key ammonia detoxifying enzymes (glutamine synthetase, glutamine dehydrogenase, alanine aminotransferase and aspartate aminotransferase) produced by the fishes' guts. Not surprisingly, the ammonia detoxifying enzymes were more active in the guts of the carnivorous fish than in the intestines of the stonerollers, which dine

on algae. In addition, the rainbow darter's ammonia levels were higher too. However, when the duo fed antibiotics to both species to knock out their gut flora – which may provide additional detoxifying enzymes to boost ammonia decontamination – they were surprised that the enzyme levels of the carnivorous rainbow darters were unaffected. In contrast, the levels of alanine aminotransferase in the stonerollers' intestines fell by 70%. 'This suggests that the carnivorous fish is not (or is less) reliant on bacterial inhabitants for ammonia detoxification', says Turner. And when she checked which bacterial species inhabited the fishes' guts, the populations differed between the two species, but both were dominated by a group of bacteria known as the Proteobacteria.

So, why are the rainbow darters less reliant on backup from their bacterial lodgers than their vegetarian stoneroller cousins, when they are likely to have a higher risk of ammonia poisoning? Bucking and Turner suspect that the risk is too great for the carnivores to depend on bacterial support. 'If the bacteria were to change, as can occur..., the fish could potentially be exposed to lethal ammonia concentrations', says Bucking, who is keen to find out more about the complex relationship between the fishes' gut bacteria and their digestion as each of them make the most of their carnivorous and vegetarian diets.

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