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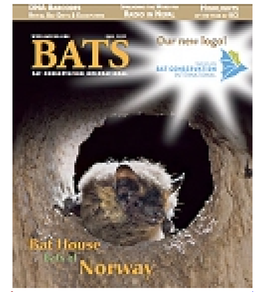

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Bats, Bugs and Barcodes
?DNA technology spotlights bats and their ecosystems
Elizabeth Clare and Robin Floyd

All through the night, bats are swooping across the skies as they feed on a wide range of insects. But they hunt mostly in secret, hidden from our view by the darkness. Figuring out what bats eat sounds like a fairly simple problem, but it is, in fact, a hot topic in the world of bat biology.

Traditionally, researchers examine bat guano under a microscope and try to visually identify insects from the bits and pieces that survive the trip through the bat's digestive tract. Identification to species level is often impossible, and many small, soft-bodied insects simply escape detection. But a new technology called "DNA barcoding" is allowing us to identify the exact species of insects eaten by bats using the DNA left in bat guano. Our research confirms the power of this approach for examining not only the behavior of bats, but also the environmental conditions in which they live and feed.

We previously reported in BATS magazine ("Food Forensics," Winter 2009) on our use of DNA barcoding in diet analysis for the first time in bats, confirming its value in identifying the diet of eastern red bats (Lasiurus borealis) in Ontario, Canada. We have since applied this relatively fast and inexpensive technique to other bat species, including the little brown myotis (Myotis lucifugus).

Traces of DNA, the raw materials of genes, can be used like fingerprints to uniquely identify a human individual, as demonstrated on countless TV programs. In much the same way, DNA can be used to distinguish one species from another. A DNA barcode is a short region of a single gene that can be read and used as a marker, or signature, for the species. The region chosen for the barcode is part of a gene called cytochrome oxidase subunit I (COI), which is present in all animals but varies by species.

Since a single cell from any animal or plant should contain its entire genome, even a tiny sample â€“ a piece of insect leg, a hair, even digested remains â€“ are usually sufficient for DNA barcoding.

But a DNA sequence by itself doesn't tell you much. You also need a reliable database that lets you match each COI sequence with the correct species. We use the Barcode of Life Data Systems (BOLD), a growing database of COI sequences from more than 100,000 species. The database includes pictures, maps, data on taxonomy and links to museums and expert information. BOLD functions as a sort of "encyclopedia of life" â€“ a resource that is both used and expanded by scientists all over the world.

Our research team from the University of Guelph, the University of Western Ontario and the Stroud Water Research Centre focused on the little brown myotis in Ontario. Results of the study were published this year in the journal Molecular Ecology. Though one of the most widespread bats in North America, this species is presently under severe threat from White-nose Syndrome.

We collected guano on sheets placed under bat roosts at three sites in southwestern Ontario â€“ two in agricultural areas and one in a forested conservation area â€“ from May to August, spanning the bats' maternity season. The samples were frozen after collection.

Later, at the University of Guelph's Biodiversity Institute of Ontario, we teased apart the guano pellets under a microscope and placed the tiny insect fragments we collected into tubes where DNA was extracted and sequenced. With the COI sequences in hand, we searched the BOLD database for matches.

We identified a total of 61 different insect species and 4 species of spiders that had been consumed by bats at these maternity roosts.

These results led to several significant observations about the bats' diet. First, many of the insects were either from the genus Chironomus (small flies called midges) or Caenis (mayflies). These two groups of insects comprised almost half of all our identifications. The remaining fragments came from a variety of insect groups including moths, beetles, caddisflies and ants.

The diet of these bats varied through the maternity season and among the three sites. Bats in the forested conservation site appeared to consume a wider variety of prey than those in agricultural areas, even though the forest was on the outskirts of Toronto, a heavily urbanized environment. This site has a small forest patch and a small pond that support many insects. The varied diet we found with these bats suggests that protecting even small forest fragments can dramatically increase bats' dietary diversity.

We also noted that at all sites, pregnant bats relied heavily on midges, but as soon as the pups were born, they switched mostly to mayflies. This might be a result of environmental changes, such as weather, that altered insect populations, or perhaps it's because the physiological demands of milk production caused the bats to change food preferences.

The most intriguing finding of the study is that the bats were mostly consuming insects that emerge from water sources â€“ and many of these aquatic species are what scientist call "environmental indicator species." These species have such specific habitat requirements that they can be used to reconstruct a predator's foraging habitat.

At one of the agricultural sites, we found identified insects in the guano that were characteristic of moving water, such as a small river or stream. The other two sites revealed insect prey that live only in still water, such as lakes or ponds. We were able to observe the foraging behavior of bats at the forest site and confirm that they were hunting over a small pond less than 330 yards (300 meters) from their roost.

In addition, many of the insects that bats were consuming have documented "pollution tolerances." The presence or absence of such insects has long been used as a key indicator of water quality at streams and lakes. The indicator species consumed at all our sampling locations indicated fair-to-good water quality, although none suggested pristine habitat.

Finally, ant species are usually good indicators of a forested habitat, and these were found only in guano from bats in the forested environment, which suggests these bats are sticking rather close to their roosts when foraging.

Our continuing research demonstrates that precise information about diet can reveal key insights into the biology of bats, and that forensic DNA research also highlights critical

links between such environmental factors as water quality and the predators at the top of the food chain.

This technology offers an exciting new method of learning about bats in a noninvasive way, and it is bringing their behavior and their ecosystems out of the dark.

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