BARCODE OF LIFE

BY SARAH KELLETT

IMAGINE WALKING THROUGH AN AMAZON RAINFOREST LOOKING FOR FROGS TO STUDY. SUDDENLY, YOU SPOT ONE: BRIGHT YELLOW AND RED. YOU'VE NEVER SEEN ONE QUITE LIKE IT, SO YOU CATCH IT, WIPE OFF A SMALL MUCOUS SAMPLE, AND PUT THE SAMPLE INTO YOUR HANDHELD DNA BARCODING DEVICE. THE FROG COULD BE A NEW SPECIES.

BEGINNINGS OF A BIG IDEA

Stroll through the grocery aisle and look at the barcodes – unique black and white patterns with 100 billion possibilities, which link products on the shelf to data in a computer. For Paul Hebert, it sparked an idea. Could species be identified the same way using DNA – the information rich sequence within every living organism? A hundred thousand species later, his dream is becoming a reality.

Paul is Scientific Director of the International Barcode of Life at the University of Guelph in Canada. He is known as the godfather of DNA barcoding – a process that links a small section of DNA to a species name. Researchers from around the world are now identifying barcodes for fish, birds, butterflies, and other animals and plants.

From 28 November to 3 December 2011, the University of Adelaide is hosting the Fourth International Barcode of Life Conference. Scientists are meeting to discuss their projects and the latest research in barcoding technology. What's it all about?

MEET PAUL HEBERT, GODFATHER OF DNA BARCODING

"I HAVE HAD A LONG TERM FASCINATION WITH INSECTS. IN FACT, MY EARLIEST RECOLLECTION IN LIFE IS COLLECTING A BUMBLEBEE AS IT VISITED A PEONY PLANT IN OUR BACKYARD. AFTER BOTTLING THE BEE, I DASHED PROUDLY TO SHOW MY MOTHER, TRIPPED ON A ROCK, BROKE THE BOTTLE, GASHED MY HAND, SAW THE BEE WING AWAY AND WAS SOON ON MY WAY TO THE HOSPITAL FOR FIVE STITCHES. DESPITE THE WAR WOUND, I REMAINED FASCINATED WITH BUGS."

HOW DNA BARCODING WORKS

Barcodes use numbers and patterns to code information, whereas DNA, which is a large molecule, uses chemicals called bases to code information. Bases are similar to each other, but have slightly different shapes. Four bases make up DNA. They're named guanine, adenine, thymine and cytosine – or G, A, T and C for short.

Different species have different sequences of DNA, which means the bases are joined together in different orders. As species evolve over time, their DNA sequences slowly mutate, picking up small changes in the code. Variations within the barcode sequence are generally small, or zero, within a species, and large between different species.

The section of DNA used for barcoding animals is a gene called cytochrome c oxidase I (COI). Different segments of DNA are used for plants and fungi. With 648 bases, the COI DNA segment is small enough to make sequencing cheap and quick, but large enough to display variations between most species.

There are some limitations to barcoding. Two species sometimes share a barcode, particularly if they have only recently evolved enough to split from being the same species. Occasionally, individuals within a species have very different barcodes, though this might suggest the existence of a new species that has not yet been identified.

THE BARCODE OF WESTERN AUSTRALIA'S KING PARROT, *ALISTERUS SCAPULARIS*. THE BASES ARE COLOURED GREEN (A), BLACK (G), BLUE (C) AND RED (T).

Credits: istockphoto.com, Barcode of Life Data Systems

CRACKING THE CODE

To collect a barcode from an animal, scientists collect a tiny sample of blood or tissue. The samples are mixed with chemicals to extract the DNA from inside the cells.

The barcode region of the DNA is copied over and over again using a process called polymerase chain reaction (PCR) amplification. The process heats and cools the DNA in a mixture of biological chemicals, working a bit like a photocopier to turn a small amount of DNA it into a large sample that can be analysed.

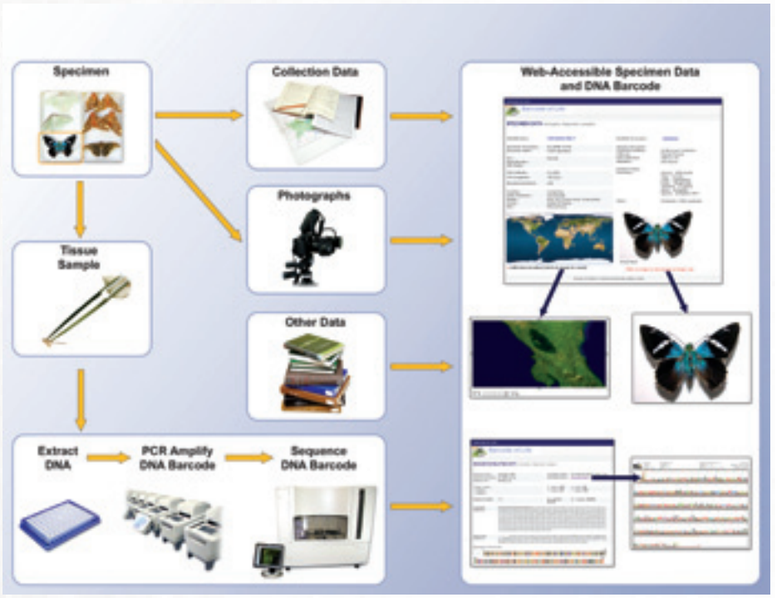
With a large sample of barcode DNA, it's time for sequencing. A machine displays which base occurs at each position, producing a series of letters, such as CCTATACCTA, which can be matched to a species.

Using DNA for identification is not new. Forensic teams may sometimes use DNA fingerprinting to catch criminals. What's different about barcoding is that it uses a standard section of DNA for all animals, making it easier for mystery samples to be quickly matched to a species.

WITHIN THE BARCODE SEQUENCE, WE HUMANS HAVE ONLY ONE OR TWO DIFFERENCES BETWEEN US, WITH ABOUT 60 DIFFERENCES BETWEEN US AND CHIMPANZEES.

BUILDING A DATABASE

Barcode sequences are stored online, in places such as the Barcode of Life Data Systems (BOLD), along with the species name, pictures and other information. The website is open to the public, so anyone can use it (www.boldsystems.org).



THE DNA BARCODE LIBRARY CONTAINS PHOTOGRAPHS, INFORMATION AND THE DNA BARCODE MATCHED TO A SPECIES NAME.

Paul believes the database could one day be accessed in the field, measuring the biodiversity around us.

"The long term goal is the automation of species identification and the development of handheld devices (like GPS) that will allow anyone to identify any organism by simply touching it [with the device]," he says.

Though it's not finished, and the machines are currently only found in laboratories, the database is already being used to identify samples, such as a piece of sushi (raw fish), to known species.

BARCODE BLITZ ON LIZARD ISLAND

Scientists are rapidly adding to the BOLD database, with projects such as FISH-BOL. This international project aims to barcode all fish species with groups set up around the globe.

In Australia, FISH-BOL plans to barcode the 4500 species of Australian fish, a goal that took a group to Lizard Island on the Great Barrier Reef off the coast of Queensland. Lizard Island houses an Australian Museum research station, and living just off shore are 10 per cent of all marine fish species. It's one of the highest diversity areas on the planet.

"We had a barcode blitz on Lizard Island," says Bob Ward, from CSIRO. "A group of us went out for a fortnight and collected fish."

The team included scientific scuba divers, taxonomists (who classify species) and geneticists (who study DNA).

LIZARD ISLAND, NEAR THE GREAT BARRIER REEF, WAS BASE CAMP FOR THE FISH-BOL BARCODING BLITZ.

Credits: International Barcode of Life, Oliver Lucanus/Belowwater.com, istockphoto.com

"During the morning and afternoon, we were collecting fish," says Bob. "The research station has some small boats, so we would go out on the reefs and snorkel". The Great Barrier Reef is a protected area, so they needed an all clear from authorities before collecting specimens and there were some species they were not allowed to catch.

A SCUBA DIVER CATCHES FISH FOR LATER DNA SAMPLING.

Taxonomists identified exactly which species had been caught, and geneticists sent a sample of DNA labelled with the species name to Canada for barcode sequencing. After two weeks at Lizard Island, they had added 380 species to the BOLD database.

SUSPECT SHARK FINS

Shark fin soup is a delicacy with a dark side. It's not so bad if fins are collected from dead sharks, but there are serious concerns if fins are cut from living sharks that are then tossed back into the ocean. Unable to swim properly, the sharks die from suffocation or are eaten.

Fins look much the same, so finding out which species a fin belongs to is virtually impossible without looking at the DNA. Bob, along with scientists from CSIRO and the University of Guelph, used DNA barcoding to identify dried shark fins confiscated from a vessel illegally fishing in Australian waters.

Only pectoral fins from the left side were used in the study, so each came from a different individual. Of 193 fins with DNA intact, barcoding detected 20 species of sharks and seven species of rays. One species, the knifetooth sawfish, is considered critically endangered. This information could help prevent species decline and overfishing.

Credits: Oliver Lucanus/Belowwater.com

SNEAKY TEA

Does the stuff in your tea bag match what's on the label? Students from Trinity School in New York wanted to find out. Katie Gamble, Rohan Kirpekar and Grace Young used equipment they bought online to investigate the DNA barcodes in herbal teas.

DO YOU KNOW WHAT'S IN YOUR TEA? DNA BARCODING CAN HELP!

More than a third of the herbal teas the students analysed contained plant species that were not listed on the package. This could be a concern for people with allergies. Their results were published in the July 2011 issue of *Nature* journal *Scientific Reports*, an achievement not to be sneezed at – many research scientists seek to be published in science journals.

"In order to analyse DNA from one type of tea, we first ground the sample material into a fine powder using a kitchen mortar and pestle," says Katie. "This helped break open the plant cell wall and release the DNA. We then did PCR to amplify the genes of special interest to us and sent the material from the PCR process to a company that could sequence our results." The genes they used were the DNA barcode sequences that are used to identify plant species.

"We received our sequenced genes the next day by email and entered that code into GenBank, the NIH's (National Institute of Health) genetic database, to find a matching species," explains Katie.

Tea bags and shark fins are just the tip of the barcoding iceberg. The International Barcode of Life plans to collect DNA barcodes from half a million species by 2015, and anyone can access the online database to find hidden species, identify mislabelled fragments or explore their local biodiversity.