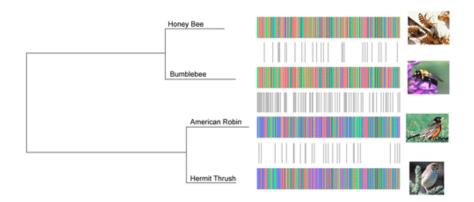


Published: 28 November 2011

# Adelaide a focus for the 'barcode of life'

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The world may contain up to 50 million species of plants and animals. But after 250 years of taxonomic research, less than two million have been named. How can we ever hope to get the job done? The solution may lie in a visionary plan to digitally barcode every species using 'bits' of DNA.



#### Credit: PloS Biology

The 4th International Barcode of Life Conference gets underway in Adelaide today, co-hosted by the University of Adelaide and the Consortium for the Barcode of Life (cBOL).

It's the largest such conference to date, with more than 450 researchers from around 60 countries debating the use of barcodes to identify crop pests, biosecurity threats, illegally sourced timber, retail fish substitutions, and even entire ecosystem communities. While its scope is huge, the science of DNA barcoding is barely eight years old.

Simply put, DNA 'barcodes' are genetic fingerprints that represent certain sequences of DNA in short gene segments found in most animals and plants. A gene known as CO1, which is found in mitochondrial DNA, is the standard barcode sequence used for animals. Its DNA varies enough to differentiate butterflies, fish, flies and other groups of animals at the genus, species and even finer levels. Two different genes – known as matK and rbcL – are used as the basis of plant barcodes.

Once the scientific world agreed to use these three bits of DNA as standard units, it triggered an international collaboration to gather DNA barcode records from five million specimens – representing at least 500 000 species – by 2015.

This collaboration – known the International Barcode of Life (iBOL) project – also aims to make its growing barcode reference library, the Barcode of Life Data Systems (BOLD), freely accessible to the wider public, including those in poorer countries lacking such a resource. Currently, more than 1.4 million specimens have been barcoded from almost 118 000 species.

Barcoding is primarily used to accurately identify species that have already been described. If a new species turns up - that is, if a specimen's barcode does not match those from already-named species – it needs to be referred to a taxonomist.



Biosecurity is benefiting from DNA barcoding. This exotic tropical nut borer, *Hypothenemus obscurus*, attacks macadamia nuts in Hawaii. Macadamia is Australia's only native plant to be widely grown as a crop around the world. Researchers have demonstrated that DNA barcodes could be used by quarantine authorities here to easily distinguish this pest species from closely related native Australian beetles.

'The concept of a global database of species-diagnostic DNA sequences is so simple, it is surprising that nobody thought of it sooner,' says Chair of the Australian Barcode of Life Network, Dr Andrew Mitchell from the Australian Museum.

'What I find truly amazing is the infectious enthusiasm of DNA barcoding's champion, Professor Paul Hebert [from Canada's Guelph University, who will be at the Adelaide conference]. He has driven iBOL through tempestuous seas since 2003.'

# 'Barcode blitz'

CSIRO's Wolf Wanjura, who is involved with the Atlas of Living Australia (ALA), points out that, once developed, protocols can be widely used by trained non-specialists. This is because the barcoding research community has developed precise protocols for running samples through the DNA barcode 'pipeline', and because genetic sequencing technology has become faster and cheaper over recent years, thanks to the Human Genome project.

'There are standard protocols to extract and work-up DNA from tissue samples to get the resulting barcode,' says Mr Wanjura. 'This offers a huge advantage in providing an alternative path to establishing an organism's identity.'

To demonstrate the speed of the technique, a group of scientists from Canada's University of Guelph worked with CSIRO, the ALA and the Australian National Insect Collection (ANIC) to carry out a 'barcode blitz' on 28 000 moth and butterfly specimens from the collection over ten weeks during 2010–11.



Credit: CSIRO

At the end of the project, led by CSIRO's John La Salle, the team had created barcodes for more than 8000 species – about 65 per cent of Australia's 10,000 known butterfly and moth species – using tissue from specimens for DNA sequencing. The barcodes were uploaded to the ALA, ANIC and BOLD databases.

# Verifying timber 'chain of custody'

The University of Adelaide's Professor Andrew Lowe is a pioneer of DNA barcoding in Australia, and Co-Chair of the Adelaide conference with Dr David Schindel from cBOL.

Rather than seeing DNA barcoding as a threat to traditional taxonomy, Prof. Lowe sees it as complementary, suited to applications that require rapid identification by non-taxonomists, such as biosecurity.

'Taxonomy and systematics is about collecting information required to identify species,' says Prof. Lowe. 'However, it may only be necessary to know whether a sample of plant or animal material – seeds, for example – is from an invasive or non-invasive species. With DNA barcoding, you can differentiate between the two: without having to wait for the seed to germinate or flower.'

The international Quarantine Barcoding Of Life project is assembling DNA barcodes to identify all invasive species – existing and potential – so that suspect material found at security checkpoints can be quickly identified.

'In South Australia alone, 1000 new plant species have been recorded over the last 20 years, one-third of which are invasive,' says Prof. Lowe. 'It's sometimes difficult for local taxonomists to identify exotic species, but now they can use DNA barcodes.'

Helping the timber industry uncover illegal product substitution is another problem being solved by DNA barcoding. Working with the Singapore-based Double Helix Tracking Initiative, Prof. Lowe used barcoding technology to investigate the chain of custody of rainforest timber harvested from natural forests in South-East Asia.



Credit: Dimas Ardian/istockphoto.com

Prof. Lowe is also applying the idea of 'metabarcoding' entire ecosystems in Australia. With funding from the Terrestrial Environment Research Network (TERN), he is establishing a national project to monitor 1000 one-hectare plots across the country.

Through the project, data from universities, museums and government agencies – such as the ALA – will be integrated into a single resource. Researchers will also take field samples from each plot to identify plant and animal material (such as pollen grains), soil microorganisms, and levels of carbon in soils and plant matter.

### The taxonomist's view

The prospect of barcoding the planet's species is daunting, so iBOL has divided its task into different 'campaigns', each focusing on different priority biological groups – for example, grasses, or moths and butterflies, or polar biodiversity.

CSIRO's Dr Bob Ward is co-chair of the international FISH-BOL campaign. He is one of a team of fish taxonomists using DNA barcoding to routinely barcode fish, shark and ray specimens deposited with the Australian National Fish Collection based at CSIRO's Hobart laboratories.

Dr Ward sees DNA barcoding as an additional and powerful tool to help taxonomists identify unknown specimens – from egg to adult – as well as undescribed species.

'There are many such fish species in Australian marine waters that taxonomists are now busily describing and naming,' he says.

'I have been assessing the extent of barcode variation within and between species. About 98 per cent of the species that we have examined can be accurately identified using the CO1 barcode – very few cannot be separated.'

DNA barcoding is proving especially effective in identifying the origin of shark fins, which command high prices in South-East Asia. This demand is contributing to the global decimation of sharks. Practices within this fishery are particularly gruesome – after sharks are 'finned', their carcasses are often dumped overboard.

Although harvesting shark fins is illegal in Australian waters, the practice still occurs. DNA barcoding recently revealed the species of origin of shark fins taken from a confiscated, illegal catch in northern Australian waters.



Credit: William White, CSIRO

'With traditional taxonomic keys, isolated shark fins are hard to identify to species level. But now we can compare the fin barcode with our reference database of barcodes from identified specimens to find the species,' says Dr Ward. 'Among the shark fins we've identified were fins from the critically endangered narrow sawfish (*Anoxypristis cuspidata*).'

Some pairs of fish species cannot be separated using the CO1 barcode – a situation Dr Ward says could be solved by establishing a secondary barcode database using a second DNA marker that would allow most of the 'difficult' species pairs to be distinguished.

'After a somewhat controversial start, [barcoding] has proved to be an extremely useful identification tool for most animal species. But barcoding, at least in Australia, still seems to be done on a shoe-string, and wider support both at institute and government levels needs to be found.

'It has huge potential, but its potential isn't yet being fully realised here.'

# **Tighter biosecurity**

Dr Mitchell is interested in the use of DNA barcoding to accelerate basic taxonomic research on moths, and to develop tools for the rapid identification of insect pests.

He is currently working with the New South Wales Department of Primary Industries and the Northern Australia Quarantine Service to barcode biting midges (*Culicoides* species) from the Australasian region. *Culicoides* carry livestock viruses, such as blue tongue.

'Our work is demonstrating that some of the important vectors [organisms that spread disease] thought to be widespread across South-East Asia are actually independent "cryptic" [difficult to distinguish]<sup>1</sup> species with more limited distributions,' he says.

'As *Culicoides* species differ widely in their vector competence [i.e. potential to carry different viruses], the implications for biosecurity are enormous.'

Dr Mitchell is excited about the possibilities of 'third-generation' DNA sequencing technologies to improve high-throughput barcoding of ecosystems.



Credit: Andrew Mitchell

In other recent developments, the application of ancient DNA methods is enabling old plant and animal specimens with potentially degraded DNA to be barcoded.

'It opens up a wealth of opportunities for museums, our storehouses of historical, current and future biodiversity information,' says Dr Mitchell.

What does he see as the biggest barrier to the take-up of DNA barcoding?

'While the public is excited by the prospects for DNA barcoding, a vocal minority of scientists remain sceptical in the face of mounting evidence,' he says. 'Ironically, those who would benefit most from barcoding, including some of the gatekeepers of biodiversity collections, are among the sceptics.'

'I'm hoping that the Adelaide conference will be a catalyst for change.'

1 For a definition see http://en.wikipedia.org/wiki/Cryptic\_species\_complex

From ECOS online http://www.ecosmagazine.com/?paper=EC11117