

DNA and Animal Classification

YEAR TEN STUDENTS



QGC

FUTUREMAKERS



**QUEENSLAND
MUSEUM NETWORK**



**Queensland
Government**

Introduction

The Molecular Identities Laboratory (MIL) at the Queensland Museum was established in 2002, and allows museum scientists to extract DNA from tissue samples to achieve a variety of outcomes. For example, scientists can use DNA sequences to help determine if they have discovered a new species. Scientists can also compare DNA sequences from different organisms and measure the number of changes (mutations) between them to infer if species are closely or distantly related.

The MIL has been used for research projects as rich and diverse as there are species to study; current and past projects have investigated Australian brush turkeys, koalas, sugar and squirrel gliders, rainbow skinks, leaf-tailed and ring-tailed geckos, artesian spring snails, sea spiders (pycnogonids), marine sponges, decapod crustaceans, scorpion fish, sea anemones, and avian and aquatic animal parasites!

As this resource has been designed to complement classroom-based teaching and learning experiences, students are assumed to have developed knowledge about the following concepts:

- Classification helps organise the diversity of life on Earth
- Species have binomial scientific names

Australian Curriculum Links

YEAR 10

Science Understanding

Biological Sciences

Transmission of heritable characteristics from one generation to the next involves DNA and genes (ACSSU184)

The theory of evolution by natural selection explains the diversity of living things and is supported by a range of scientific evidence (ACSSU185)

Science as a Human Endeavour

Nature and Development of Science

Advances in scientific understanding often rely on technological advances and are often linked to scientific discoveries (ACSHE192)

Future Makers is an innovative partnership between Queensland Museum Network and Shell's QGC project aiming to increase awareness and understanding of the value of science, technology, engineering and maths (STEM) education and skills in Queensland.

This partnership aims to engage and inspire people with the wonder of science, and increase the participation and performance of students in STEM-related subjects and careers — creating a highly capable workforce for the future.

© Queensland Museum. The images included in this teaching resource may be used for non-commercial, educational and private study purposes. They may not be reproduced for any other purpose, in any other form, without the permission of the Queensland Museum.

What is DNA?

DNA, or deoxyribonucleic acid, is the molecule of inheritance found inside the nucleus of cells.

DNA consists of several components:

- four bases (adenine or A, guanine or G, cytosine or C, and thymine or T).
These bases bond to each other, A with T, and G with C, to form the core of the molecule.
- a “backbone” made of sugar (called deoxyribose) and phosphate groups.

Together these components make the structure of DNA, which has the iconic double helix shape shown in Figure 1.

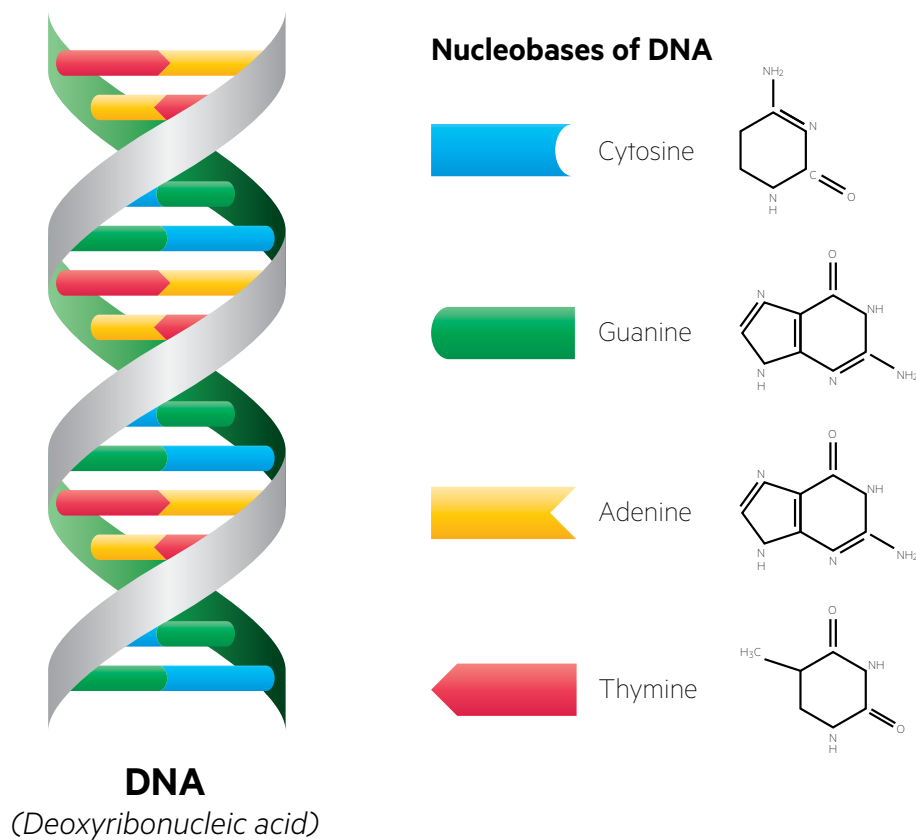


Figure 1: The structure of DNA

DNA is often referred to as the blueprint of life; it contains the instructions for how to make a living organism. How does DNA actually do that?

DNA tells the cell's components to make proteins; the order of the four DNA bases (A, T, G and C) determines which proteins are made. This is important, because proteins are essential for the building and repair of all cells and tissues, and are used to make enzymes, hormones, and many other biological molecules. For example, keratin, a structural protein, is found in skin, hair, teeth and nails; pepsin is a digestive protein that helps break down food; and haemoglobin is a respiratory protein that allows red blood cells to carry oxygen.

DNA is studied by many different kinds of scientists, including geneticists, medical researchers and taxonomists. When scientists study DNA sequences, they look at a series of letters representing the four bases, as shown below.



The above sequence could represent DNA from a human, a jellyfish, or a bacterium; all living things use G, T, A and C as their DNA bases.

The existence of the same four DNA bases in all living things is one of the many lines of evidence that suggest that all life evolved from one common ancestor.

DNA in Species Identification

Evolutionary theory states that inherited characters (like DNA sequences) change over time. Therefore, closely related organisms will have more similar DNA sequences than distantly related organisms, and the longer two species have been separated from a common ancestor, the more their DNA sequences will differ. For example, human DNA sequences are about 97% identical to chimp DNA sequences¹, while human DNA sequences are about 85% identical to mouse DNA sequences².

Closely related organisms may also have a more similar appearance, or morphology, than distantly related organisms. For example, humans and chimps share more physical characteristics (e.g. bipedalism, opposable thumbs) than humans and mice. However, it can be difficult to determine how closely related organisms are. Some organisms (like sponges, see below) don't have many visible features, while others (like coral) can change their appearance based on environmental conditions. In these cases, scientists can use DNA to help clarify the relationships between organisms. Scientists can do this by using DNA sequences to create a phylogenetic tree.

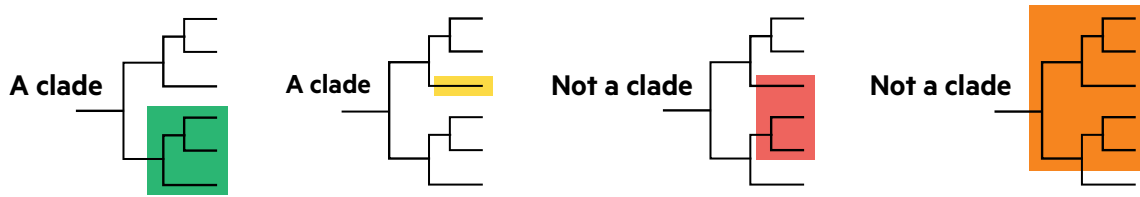


Two different sponge species (*Clathria kylista*, left, and *C. vulpina*, right). Images: John Hooper, QM.

¹Varki, A & Altheide, T. K. (2006). Comparing the human and chimpanzee genomes: Searching for needles in a haystack. *Genome Research* 15: 1746-58.

²Makalowski, W., & Boguski, M. S. (1998). Evolutionary parameters of the transcribed mammalian genome: An analysis of 2,820 orthologous rodent and human sequences. *PNAS* 95: 9407-9412.

Phylogenetic trees are branching diagrams that represent how closely related organisms are. The most closely related organisms are found in the same clade, or branch; more distantly related organisms are found in different clades.



Different types of data can be used to create phylogenetic trees; if DNA sequences are used, organisms with the most similar DNA sequences are grouped together.

An example phylogenetic tree of vertebrates is shown below (Figure 2). Groups of animals that are closely related, like birds and crocodiles, are found in the same clade. Animals that are not closely related, like salamanders and marsupials, are found in different clades.

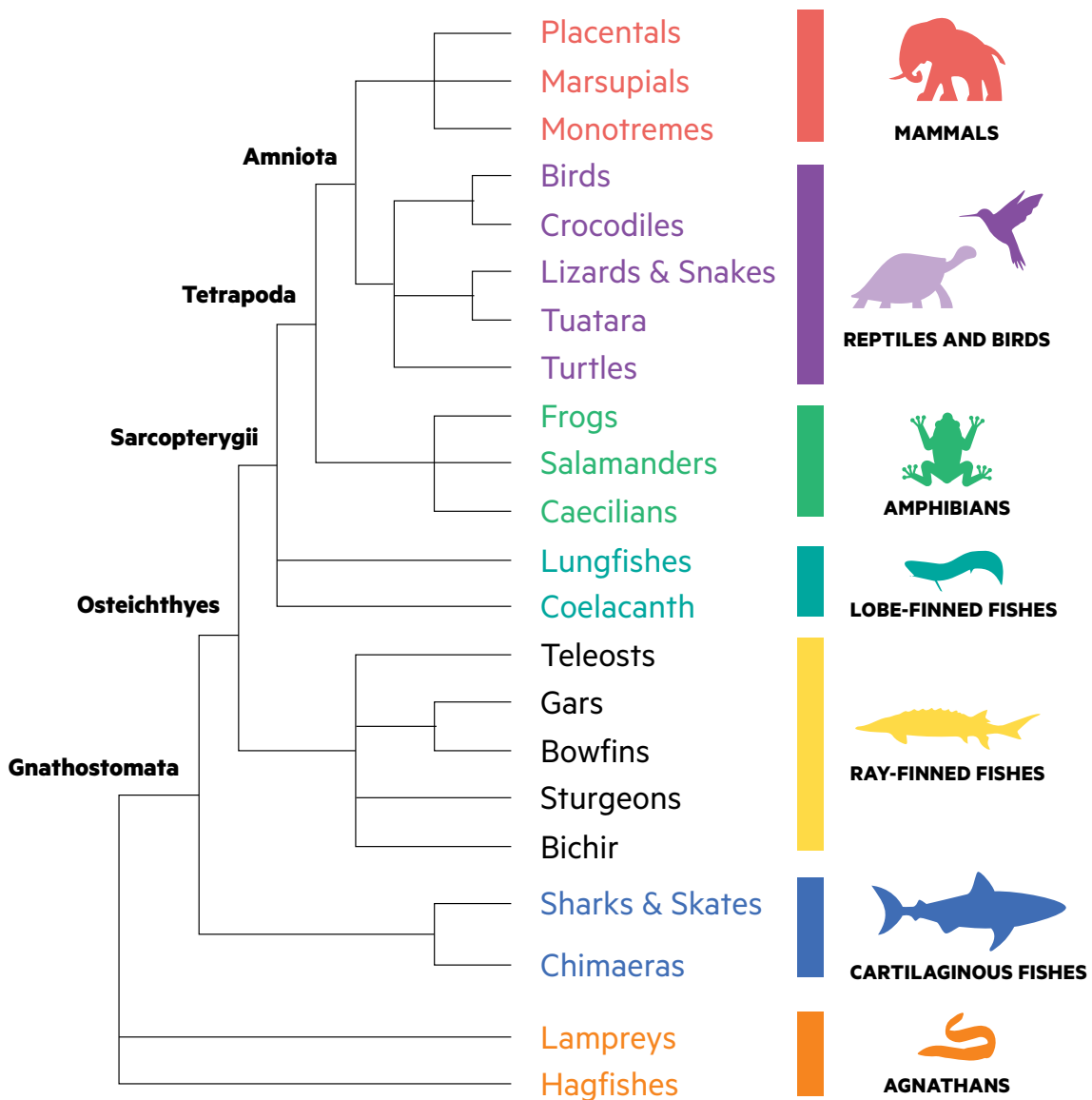


Figure 2. A phylogenetic tree of vertebrates, showing the evolutionary relationships between mammals, birds, reptiles, amphibians and fishes.

There are four DNA exercises on the following pages that demonstrate how DNA can help identify animals. In these exercises you will examine the DNA sequences of two butterflies, and use your knowledge of DNA to determine how closely related the butterfly specimens are.

Please note that the DNA sequences in each exercise are 18 base pairs long, and would only represent a tiny fraction of a real butterfly genome, which can be hundreds of millions of base pairs in length. However, even small fragments like these can help provide clues about the identity of a specimen. (The DNA sequences on pages 7-10 have been created for this document, and do not reflect real data.)



Northern Birdwing, *Ornithoptera priamus*. Image: Bruce Cowell, QM.

Butterfly images on pages 7-10: Jeff Wright, QM.

Butterfly Identification

Exercise 1



A researcher extracts DNA from two butterflies (shown above). Do you think these two butterflies belong to the same species? Why or why not?



Butterfly Identification

Exercise 2



A researcher extracts DNA from two butterflies (shown above). Do you think these two butterflies belong to the same species? Why or why not?



Butterfly Identification

Exercise 3



A researcher extracts DNA from two butterflies (shown above). Do you think these two butterflies belong to the same species? Why or why not?



Butterfly Identification

Exercise 4



A researcher extracts DNA from two butterflies (shown above). Do you think these two butterflies belong to the same species? Why or why not?



Cryptic Species: Where Morphology & DNA Diverge

Individuals that look similar or identical may not be able to interbreed, and may therefore represent members of different species. When separate species are visually indistinguishable from each other, they are known as cryptic species. DNA sequences help scientists discover cryptic species all the time. What other lines of evidence might help us discover cryptic species?

Species of litter skinks in the genus *Lygisaurus* are found in northeastern Australia, and are known for their extreme morphological conservatism relative to many other lizard genera. (Morphological conservatism means that individuals show little or no change in their physical appearance, even though their DNA sequences have changed significantly.)



Lygisaurus foliorum (top) and *Lygisaurus aeratus* (bottom) show morphological conservatism. Images: Jeff Wright and Gary Cranitch, QM.

Recent studies on litter skinks have revealed that cryptic speciation is widespread. One of these studies was done at the Queensland Museum on the species *L. aeratus* (formerly *Carlia aerata*)³. Prior to the Queensland Museum research, *L. aeratus* was thought to have a wide distribution throughout north-eastern Queensland (Figure 3, next page). Colour pattern differences had been observed among populations throughout the range, and were thought to simply reflect colour differences between the sexes, i.e. sexual dimorphism.

³ Couper, P. J., Worthington Wilmer, J., Roberts, L., Amey, A. P., and Zug, G. R. (2005). Skinks currently assigned to *Carlia aerata* (Scincidae: Lygosominae) of north-eastern Queensland: a preliminary study of cryptic diversity and two new species. *Australian Journal of Zoology* 53: 35-49.

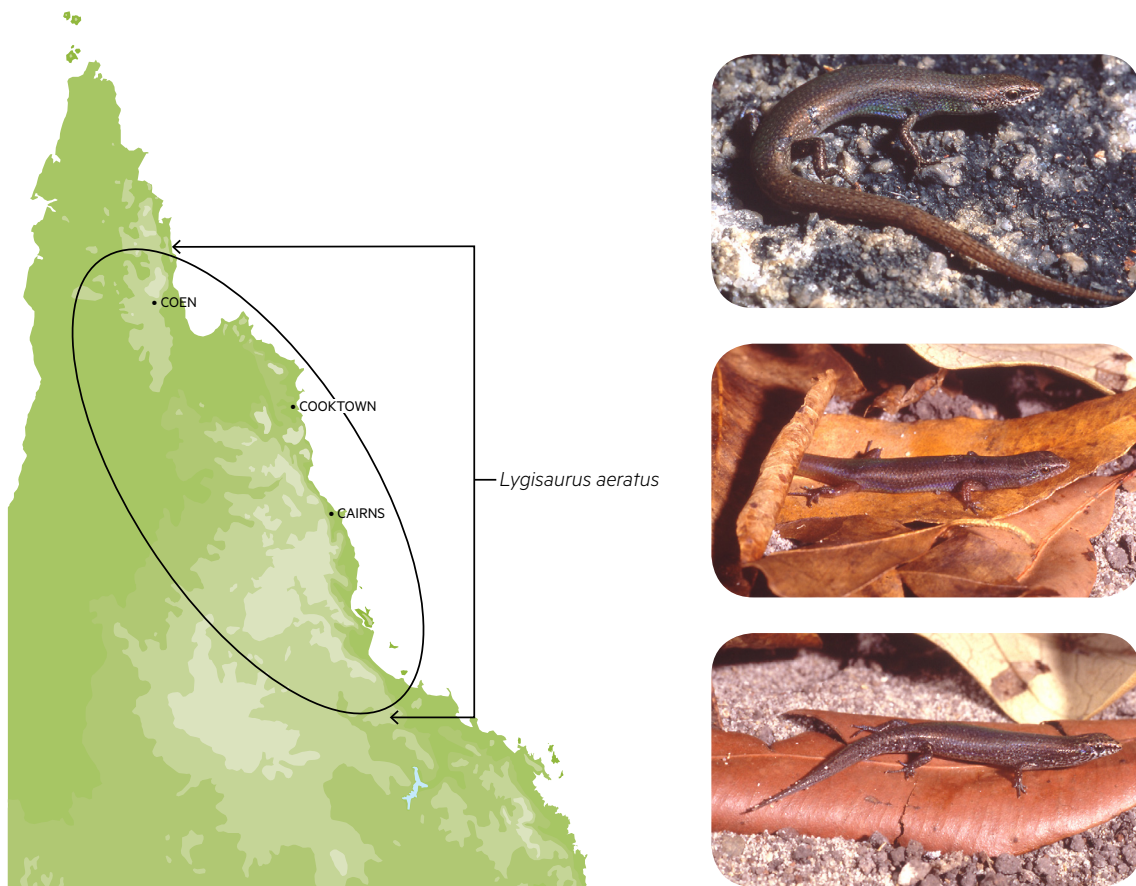


Figure 3. The distribution of *Lygisaurus aeratus* before 2005, based on available data. Images on the right show different *L. aeratus* specimens from across this range. Images: Gary Cranitch, QM, and Colin Dollery.

To investigate these colour pattern differences, Queensland Museum scientists collected tissue samples from *L. aeratus* skins from many different locations in Queensland.



Queensland Museum herpetologist Dr Patrick Couper in the field. Image: Jeanette Covacevich.

The Queensland Museum geneticist (Dr Jessica Worthington Wilmer) processed the skink tissue samples and extracted DNA from them. The resulting DNA sequences were then used to construct a phylogenetic tree of different *Lygisaurus* species (Figure 4), which suggested that the colour pattern differences actually represented three very distinct species, two of which were new to science.



Queensland Museum geneticist Dr Worthington Wilmer works on tissue samples in the lab. Image: Peter Waddington, QM.

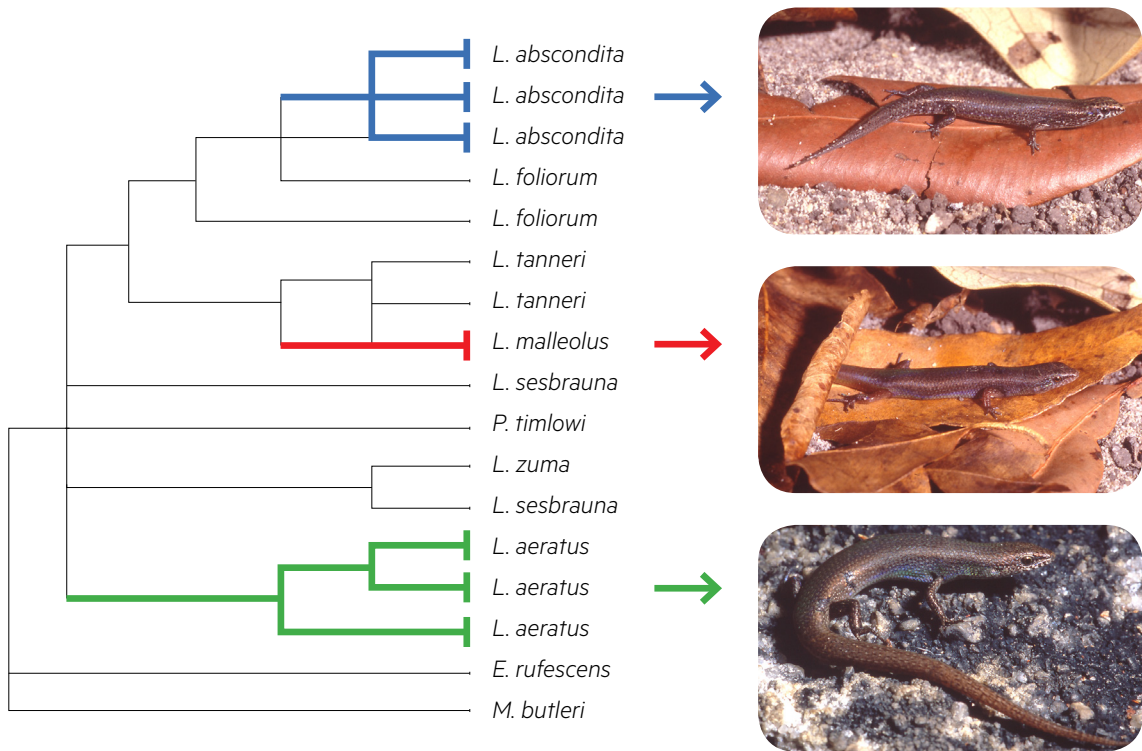


Figure 4. A phylogenetic tree for skinks from the genus *Lygisaurus* based on data from Couper *et al.*, (2005). The species in blue, red and green (shown with corresponding skinks to the right) were all originally thought to be *Lygisaurus aeratus* based on their appearance. Images: Gary Cranitch, QM, and Colin Dollery.

1. Why does this tree suggest the existence of two new species (*L. malleolus* and *L. abscondita*)?

2. Draw or explain how this tree would look if *L. malleolus* and *L. abscondita* specimens were actually the closest relatives of *L. aeratus*.

3. What might have caused these skink populations to develop into different species?

4. How might this study (and others like it) affect estimates of local biodiversity and conservation efforts?

This research changed our understanding of the species *Lygisaurus aeratus*. Instead of having a single species with a wide distribution, we now have a completely different scenario: three species, each with very different range distributions (Figure 5).

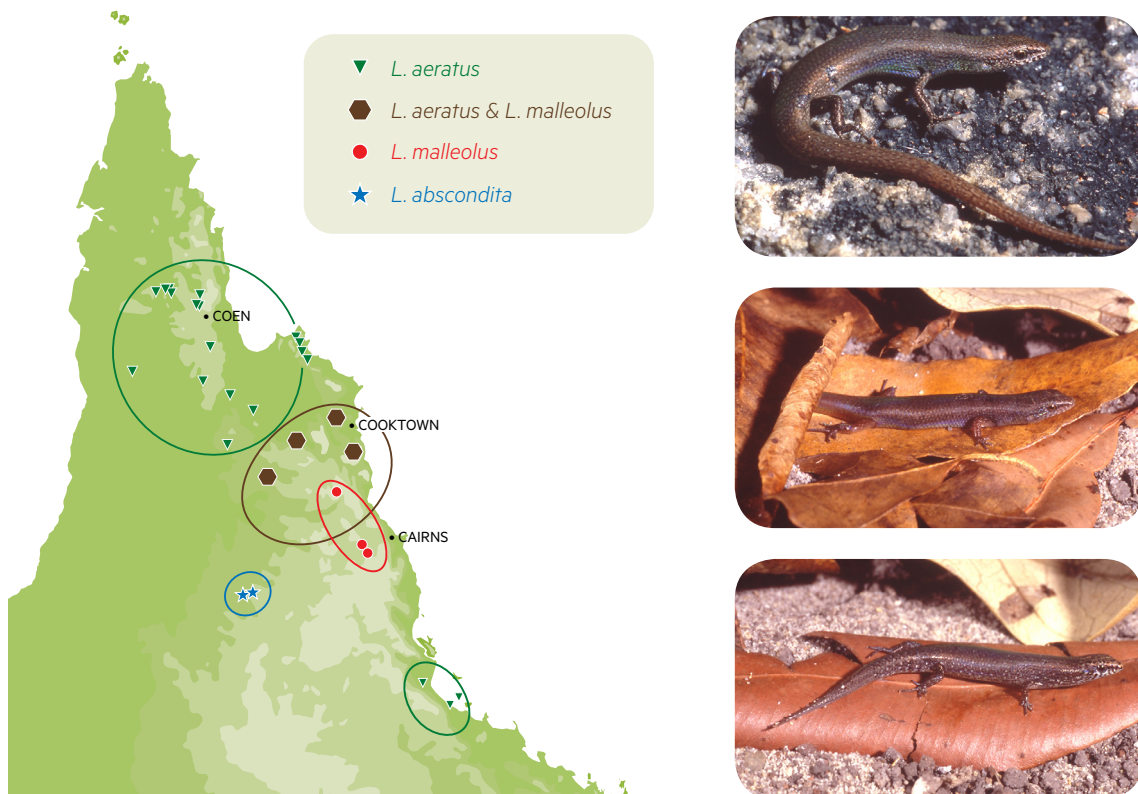


Figure 5. The updated distribution of the three species, based on data from Couper *et al.*, (2005). The brown hexagons represent sites where both *L. aeratus* and *L. malleolus* were found. Images: Gary Cranitch, QM, and Colin Dollery.

What's in a name?

Species names might seem like official titles that don't change, but actually the names of species change all the time! For example, the skink research discussed in this document was published in 2005, and at that time the species in the study were all in the genus *Carlia*. Since 2005, expanded analyses (more species, more data) by other scientists resulted in all *Carlia* species being reclassified into the genus *Lygisaurus*.

This is one example of how taxonomic classifications and species names are not static. Instead they are constantly evolving as more sophisticated technology enables new and better resolution of data.

Extension Activity

Scientific research is intimately linked to advances in technology. For example, scientists were unable to obtain DNA sequences from organisms on a large scale until the 1980s, with the invention of the polymerase chain reaction (PCR). PCR is a technique that allows scientists to quickly and cheaply make copies of any region of DNA from any organism. PCR is now used every day in labs around the world, and underpins all of modern biology research.

Research some new and emerging technologies in biology. What do you think will be the next big breakthrough?

More about the Queensland Museum experts

You can watch a video of the Queensland Museum geneticist, Dr Jessica Worthington Wilmer, and see other Queensland Museum experts at the link below:

<https://learning.qm.qld.gov.au/>

Butterfly Identification

Exercise 1



A researcher extracts DNA from two butterflies (shown above).

These two specimens look the same, and their DNA sequences are identical. These data support the hypothesis that these two specimens belong to the same species. **These two specimens are both Blue Triangles (*Graphium sarpedon*).**



Butterfly Identification

Exercise 2



A researcher extracts DNA from two butterflies (shown above).

These two specimens look very different, and their DNA sequences differ significantly (by 4 bases out of 18, or 22%). The DNA sequences of closely related species often only differ by a few percent; recall the example of humans and chimps given on page 4. Therefore, these data strongly support the hypothesis that these two specimens belong to different species. **The top specimen is a Blue Triangle (*Graphium sarpedon*), and the bottom specimen is an Orange Dart (*Suniana sunias*).**



Butterfly Identification

Exercise 3



A researcher extracts DNA from two butterflies (shown above).

Although these two specimens look similar, their DNA sequences differ significantly, as in the previous exercise (by 4 bases out of 18, or 22%). The genetic data strongly support the hypothesis that these two specimens belong to different species. Students can discuss reasons why the morphology of these two specimens might be so similar, even if they are different species. (The butterflies on this page are known as skippers. Skipper species are notoriously hard to identify because they look so similar to each other. Characteristics of wing veins, scales and genitalia are needed to identify species.) **The top specimen is an Orange Dart (*Suniana sunias*), and the bottom specimen is an Orange Palmdart (*Cepherenes augiades*).**



Butterfly Identification

Exercise 4



A researcher extracts DNA from two butterflies (shown above).

These two specimens have different colouration, although their wing shapes and body shapes are similar, and their DNA sequences are identical. In this case the DNA evidence strongly supports the hypothesis that these two specimens belong to the same species. Why do they have such different colouration? The top photograph shows a male Orchard Swallowtail, the bottom a female Orchard Swallowtail. This sexual dimorphism is just one example of how appearances can be misleading when trying to identify organisms, and shows how DNA can help by providing a separate line of evidence. **These two specimens are both Orchard Swallowtails (*Papilio aegeus*).**



It's complicated



Unlike these four simplified examples, determining whether individuals belong to the same or different species in the real world is a lot more complicated! The boundaries between species are determined by assessing differences across a range of criteria, including morphological, genetic, behavioural and ecological characters. Each of those variables may provide conflicting results, and the degree of difference in characters which constitutes a species boundary is not always easy to quantify.

“What is a species?” is a scientific question that does not have a single hard and fast answer; rather there are multiple operating concepts and definitions. Furthermore, those definitions vary widely among taxonomic groups. In short, biology is challenging because at times it is messy, which of course is part of its appeal!