For many years, the decline of frogs in various parts of the world has puzzled conservationists. A breakthrough came in 1996 when scientists isolated a new kind of fungus that infects and potentially kills frogs. Western Australian research now under way is beginning to answer some initial questions about the fungus and its impact on our unique frogs.

In pursuit of the frog fungus

by Ken Aplin and Peter Kirkpatrick

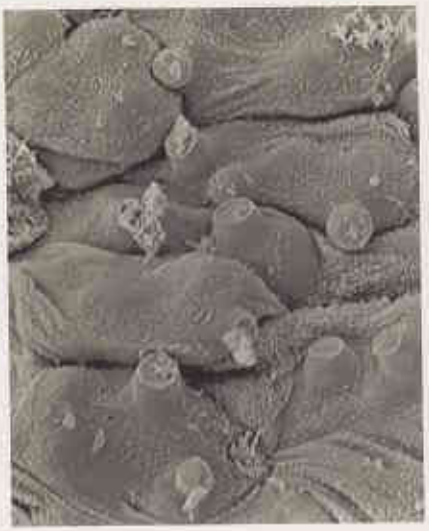
Frog fungus? Never heard of it? To be honest, neither had we before August 1998. And neither had the rest of the world prior to 1996. But work over the past two years in Western Australia has already led to a significant increase in our understanding of this newly identified and potentially harmful disease of amphibians.

Frog fungus (*Batrachochytrium dendrobatidis*) is a microscopic fungus that lives on frogs, or more correctly, as a parasite within the skin of frogs and other amphibians. It belongs to an ancient group of funguses called the Chytridiales (or 'chytrids' for short—pronounced ki-trids). These tiny organisms usually exist as free-living single-cell organisms, most often in water-bodies or in soil. Although some are parasitic on algae and aquatic invertebrates, the frog fungus is the only member of the group known to live and feed on a vertebrate host.

DISCOVERY OF THE FUNGUS

Although the frog fungus is now known from nearly every continent, it was completely unknown to science until 1996. It was finally identified as a new 'pathogen' (agent of disease) by two teams of scientists working independently, one at James Cook University in Townsville, Queensland, and the other at the National Zoological Park in Washington DC, USA. Both groups of researchers were investigating the mysterious deaths of large numbers of frogs living in apparently uncontaminated, healthy environments. In Queensland, this was happening in pristine rainforests in the World Heritage Wet Tropics Area. In America, it was happening in zoos with well-maintained and precious captive frog populations.

The breakthrough came only after several years of intensive research in each laboratory. Both teams followed many different leads, but eventually narrowed their search down to a mystery organism that appeared to be confined to the outermost layer of the skin. After comparing notes and with the help of mycologist Dr Joyce Longcore, this organism was identified as a chytrid fungus. The next step was to isolate the fungus from sick frogs and use it to experimentally infect healthy frogs. These test animals soon became infected with the fungus and developed the same clinical symptoms including lethargy and muscle spasms, leading to death. The tests established the link between the skin parasite and the disease symptoms, at least under laboratory conditions. Dr Longcore subsequently gave the fungus its near



Previous page
Motorbike frog (*Litoria moorei*).
Photo – Jiri Lochman

Left: A close-up of frog skin, heavily infected with frog fungus. The circular bodies are 'discharge tubes' through which new spores are released into the environment.
Photo – Lee Berger

Below: The motorbike frog, seen here in another colour variation, is a common victim of the frog fungus.
Photo – Babs & Bert Wells/CALM

Right: Western banjo frog is susceptible to the frog fungus, but shows no evidence of widespread decline.
Photo – Babs & Bert Wells/CALM

Below right: Adult moaning frogs rarely enter water and infection rates are lower than in many other species.
Photo – Brad Maryan

unpronounceable name, which means 'frog chytrid of dendrobatid frogs', dendrobatids being the family of poison arrow frogs, common victims of the fungus in American zoos!

In the five years since its discovery, the frog fungus has been recorded from many parts of the world. The current list, maintained on a website by Associate Professor Rick Speare of James Cook University, includes more than 90 different amphibian species from North, Central and South America, Europe, Africa, New Zealand and Australia.

Although the frog fungus was unknown until 1996, earlier records can be detected by studying historical specimens stored in various museum collections around the world. By this means, infections have already been recorded as far back as 1978 in eastern Australia and to the early 1970s in several parts of North America.

SUPPORTERS AND SCEPTICS

The discovery of the frog fungus was heralded by many researchers as a major breakthrough in the search for a common factor linking frog declines in various parts of the world. However, other researchers remained sceptical about its significance. Some expressed doubts that any wildlife disease could have such pronounced and widespread impacts, unless the affected populations were already compromised by some other stress-inducing factor such as environmental contamination or changes in global climate. Other researchers were dubious about whether the laboratory results accurately reflected conditions in the field—a common criticism of laboratory tests of this kind.

Nor could researchers agree on the reason for the differences in timing of chytrid disease outbreaks in different areas. Was the frog fungus spreading around the world, wreaking havoc on amphibian faunas with little or no resistance to the disease? Or was it a naturally widespread organism, which only emerged as a problem 'disease' when the relationship between host and parasite altered, perhaps because of environmental change or lowered immunity? Many examples of both kinds of 'emergent infectious diseases' can be cited from human history: for example, the spread of influenza into the New World in the sixteenth century and modern outbreaks of cholera following natural disasters and disruption of water supplies.

Like many scientific breakthroughs, the discovery of the frog fungus has raised many more questions than it has answered!

FUNGUS FOUND IN WA

The first evidence of the frog fungus in WA came in 1998 from a recently dead motorbike frog (*Litoria moorei*) picked up on The University of Western Australia (UWA) campus in Nedlands by a concerned zoology student. Diagnosis was performed by Lee Berger, formerly of the James Cook team, but by then based at the CSIRO Animal Health Laboratory in Geelong, Victoria.

The worrying news spread quickly. Ken Aplin appealed for help in locating sick or recently dead frogs, both through the news media and direct to several thousand dedicated members of the Western Australian Museum's Alcoa

Frog Watch. Within days, specimens were coming in from throughout the metropolitan area and beyond.

An initial sample of 70 or so sick and dead specimens was sent interstate for processing and diagnosis. The results were not good news—positive infections at various localities around Perth and as far away as Witchcliffe (near Margaret River), Manjimup and Albany. Moreover, fungal infections were recorded in at least four species of frogs. A thousand and one questions ran through our minds. Was the fungus recently introduced? Or had it always been here? Would some species be more susceptible than others and, if so, which ones?

LOCAL STUDIES

In 1999, we began a detailed study of the frog fungus in WA. This was made possible by funding from the Department of Conservation and Land Management (CALM) and the World Wide Fund for Nature Australia Frogs Program, and with training in diagnostic methods by Lee Berger and Rick Speare. Curtin University provided laboratory facilities. Additional work was undertaken by Zoe Car, an Honours student in Zoology at UWA.

Our combined studies aimed to answer five basic questions. Firstly, how widespread was the zone of infection? Secondly, how long had chytrids been present in WA? Thirdly, was there any seasonal or geographic pattern in infection rates? Fourthly, which species were being infected? And lastly, which of the infected species were showing clinical symptoms of ill-health including death? These questions were designed to provide a solid foundation for future management of the frog fungus disease in WA.

To address the first four questions, large numbers of specimens from the Museum's collection would need to be examined, preferably covering a large geographic area, a wide range of time periods and a diverse range of species. However, Museum specimens alone would not provide an answer to the last question. For this, many more sick and dead frogs would be needed to compare with the 'healthy' samples typically collected and deposited in museums. Throughout the study, repeated calls for help were issued, with good response, especially from many Alcoa Frog Watch members who took special care to monitor the health of their garden frog populations. Many specimens also came in from regional CALM offices.

After two years, we had examined more than 4,000 specimens, covering a 50-year period and representing 28 different species. Most of this material came from the south-western corner of the State—thus providing the most detailed study of a regional zone of frog fungus infection anywhere in the world.

ANSWERING QUESTIONS

Our studies answered many of our original questions. We now know that a broad zone of infection is present across the south-west corner of the State, from just north of Geraldton, south to Augusta and east to Esperance, extending inland nearly as far as Southern Cross. This doesn't mean that every frog population in the south-west is necessarily infected at any one time. However, it does mean that no site in this region can be thought of as a 'fungus free zone' in any long-term sense.

We also know that there is at least one isolated infection point on the Gibb River Road in the eastern Kimberley. This is disturbing, as there was previously no evidence of the frog fungus from anywhere in north-western Australia, although infections are known from northern Queensland.

Initial historical studies by Zoe Car produced some stunning evidence of frog fungus infections in the Albany region back as early as 1985! This led her to speculate that the fungus might in fact be native to the south-west region. However, subsequent testing of more than 1,000 samples from the period 1950–1984 failed to find any earlier cases. Rather, our more detailed study appears to track the fungus spreading outwards from an initial outbreak in the Albany region, reaching

Perth in 1989, Esperance by 1992 and Geraldton by 1998. These results clearly favour the notion of an introduction and rapid spread of the fungus, but this picture could change with further testing.

We found evidence of chytrid infection in every south-western frog species that we sampled intensively, a total of 17 in all. Quite possibly, the fungus can infect all frogs, although all may not develop any other symptoms or suffer any disease. Not surprisingly, infection rates in different frog species reflect their contrasting lifestyles. As a rule, frogs that spend more time in or near water, such as the motorbike frog, had infection rates three or four times higher than those, like the moaning frog (*Heleioporus eyrei*), that spend most of their life on land.

Four species of frogs dominated the 'sick and dead' sample brought in by members of the public. These were the two local tree frogs (the motorbike frog and slender tree trog, *Litoria adelaidensis*), the moaning frog and the banjo frog (*Limnodynastes dorsalis*). In each species, the chytrid infection rate was at least three times higher in this sample than in the corresponding 'apparently healthy and active' samples. This is strong evidence that fungal infection does indeed lead to ill-health and death of at least these species of frogs under natural conditions.

Given this conclusion, it was disturbing to find no evidence in our data that infection rates had fallen since 1985, hence no sign that local frogs were becoming more resistant to infection. Similarly, the fact that we found such high rates of infection in the 'sick and dead' sample also made it unlikely that local frogs had developed any strong resistance to clinical disease over this period.

On a more positive note, the intensity of fungal outbreaks appeared to differ from year to year, possibly with more 'good' years than 'bad' years so far. Taking only samples collected around Perth, we compared seasonal patterns for each year over the past decade. The graphs show a consistent seasonal trend in infection rates, beginning with values of one to five per cent during the dry, hot months of summer and autumn, climbing to around 10 per cent through winter, and peaking at between 20 and 50 per cent in spring and early summer, before rapidly dropping back to the summer low. Between years, the spring to early summer 'peak' has varied substantially in severity. During the 1991–92 and 1998–99 periods, every second frog around Perth was probably infected! (Significantly, some Alcoa Frog Watch members reported a 50 per cent mortality among their garden frog populations through winter to spring 1999.) However, between these periods, infection levels were considerably lower and there may have been lower rates of sickness and death among local frogs.

An unexpected result of our study was that infection rates did not seem to be influenced by environmental quality. Rates of infection were just as high in relatively undisturbed bushland habitats, away from strong human influence, as they were at some of the most disturbed and contaminated localities in the Perth metropolitan area.

PAST AND FUTURE

The really big unknowns are whether or not the frog fungus has caused any lasting declines in Western

Right: Slender tree frog (*Litoria adelaidensis*).
Photo – Babs & Bert Wells/CALM

Right: The orange-bellied frog has shown no evidence of decline, but is perhaps 'vulnerable' to disease due to its small geographic range.
Photo – Grant Wardell-Johnson/CALM

Below right: Kids netting for tadpoles—an age-old pleasure. Hopefully, future generations will continue to enjoy the experience of living together with frogs.
Photo – Ken Aplin

Australian frogs so far, and whether or not it is likely to in the future. As far as we know, WA has so far avoided the worst impacts of the frog fungus. Along the eastern seaboard of Australia, about eight frog species have become extinct over the past two decades and others are now highly threatened. Almost all of these are from high altitude areas. In the south-west region, we can be fairly confident that no frog species has become extinct or suffered a catastrophic decline over the past 15 years—perhaps the lack of high country in the south-west has somehow limited the impact of the disease? However, we cannot be so sure about other parts of the State, especially the infrequently visited Kimberley region. And have there been more subtle changes, even in the south-west?

Over the past decade, CALM staff and UWA researchers have accurately monitored populations of the white-bellied and orange-bellied frogs (*Geocrinia alba* and *G. vitellina*), both of which are restricted to small natural ranges in the Margaret River area (see 'Frogs: Value in Variety', *LANDSCOPE*, Spring 1994). The white-bellied frog is declining and is listed as 'critically endangered'. The orange-bellied frog is not, and is listed as 'vulnerable', even though there is evidence that the species was infected during 1996. This may be evidence that some species are more resistant to the fungus than others, but the picture is not clear.

Unfortunately, our work has shown that at least some groups of south-western frogs are highly susceptible to frog fungus disease. To what extent are these species under threat? The short answer is that we don't know. In many areas, populations of frogs have indeed fallen dramatically over the past one or two decades, but at present we are unable to say whether this has been caused by frog fungus disease or by one or more other factors, such as habitat degradation or our growing dependence on potentially harmful environmental chemicals. We believe in being cautious and vigilant. Our studies of the frog fungus will continue.

WHAT YOU CAN DO

After reading this article, you may want to know how you can help our frogs. To begin with, you can learn about frogs and help to raise awareness of disease and other threats to frogs. You can also help to identify major outbreaks and 'epidemic' years by collecting sick and dead frogs (contact the Museum to find out how). Another way to help is to become involved in frog conservation through assisting with landcare work. And we can all make a small but important contribution by looking after our own backyard, helping to make the suburban environment more frog-friendly overall (see 'Frogs in the Garden', *LANDSCOPE*, Winter 1993). By working together, we can help to make the State a better place for frogs.

Ken Aplin is Coordinator of the Western Australian Museum's Alcoa Frog Watch program. He can be contacted on (08) 9427 2826 or by email (frogwatch@museum.wa.gov.au).

Peter Kirkpatrick is a Curtin University Environmental Science graduate who spent 1999 working at the Western Australian Museum on the frog fungus. He is currently enrolled in Veterinary Science at Murdoch University, but continues his frog disease work part time with the Museum. He can be contacted by email (frogwatch@museum.wa.gov.au).

058605

Winner of the 1998 Alex Harris Medal for excellence in science and environment reporting.

LANDSCOPE

VOLUME SIXTEEN, NUMBER 3, AUTUMN 2001

Armed with sketch pad, pencils, pens and paints, an intrepid group of artists set off on a brand new LANDSCOPE *expedition. See 'Awash with Colour' on page 28.*

Four more conservation reserves now offer greater protection to areas in and around the Mitchell Plateau. See 'Parks of the Plateau' on page 48.

Most of us only know of the exotic pest ants that invade our kitchens. But what of the great Australian ants? See page 23.

Ningaloo Marine Park and Cape Range National Park lie side by side in our north-west corner. Read about how they are managed on page 17.

Scientists continue to develop ways to locate, track and trap animals for research. See 'Tools of the Trade' on page 41.

COVER

For many years, the decline of frogs in various parts of the world has puzzled conservationists. A breakthrough came in 1996 when scientists isolated a new kind of fungus that infects and may kill frogs. Western Australian research now under way is beginning to answer some initial questions about the fungus and its impact on our unique frogs. See 'In Pursuit of the Frog Fungus' on page 10.

Cover illustration by Philippa Nikulinsky

FEATURES

REGULARS

Executive Editor: Ron Kawalilak
Editors: David Gough and Carolyn Thomson-Dans
Bush Telegraph Editor: Verna Costello
Advertising copy and editorial assistance: Caris Bailey
Scientific/technical advice: Andrew Burbidge, Chris Simpson, Paul Jones and staff of CALMScience Division
Design and production: Tiffany Aberin, Maria Duthie, Gooitzen van der Meer
Illustration: Gooitzen van der Meer
Cartography: Promaco Geodraft
Marketing: Estelle de San Miguel ☎ (08) 9334 0296 Fax: (08) 9334 0498
Subscription enquiries: ☎ (08) 9334 0481 or (08) 9334 0437
Colour Separation by Colourbox Digital
Printed in Western Australia by Lamb Print

© ISSN 0815-4465. All material copyright. No part of the contents of the publication may be reproduced without the consent of the publishers.

Please do not send unsolicited material to *LANDSCOPE*, but feel free to telephone the Editor.

Visit NatureBase at http://www.naturebase.net

Published by the Department of Conservation and Land Management, Dick Perry Avenue, Kensington, Western Australia

DEPARTMENT OF Conservation AND LAND MANAGEMENT
Conserving the nature of WA