Statement of Intent

2: Infection of frogs by amphibian chytrid causing the disease chytridiomycosis

Department of
Environment, Climate Change and Water NSW

Cover photo:
Spotted tree frog (*Litoria spenceri*) being swabbed (Stuart Cohen, DECC)

Published by:
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ISBN 978 1 74232 367 1
DECCW 2009/479
September 2009

Printed on recycled paper

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1. Introduction

Chytridiomycosis is an infectious disease that affects amphibians worldwide. The disease is caused by a fungus known as the amphibian chytrid fungus or *Batrachochytrium dendrobatidis* (Longcore et al. 1999).

Of the nearly 6000 amphibian species known worldwide, over 43 per cent are currently in a state of decline and over 32 per cent are currently at risk of going extinct. Although there are many factors contributing to these declines, chytridiomycosis has been identified as a major cause, driving amphibian species towards extinction at a rate unprecedented in any taxonomic group in human history (Gascon et al. 2007; Skerratt et al. 2007). In 2002, chytridiomycosis was placed on the Wildlife Diseases List by the World Organisation for Animal Health as a disease of global significance (DEH 2006b) and declared a notifiable disease in 2008.

In Australia, chytridiomycosis has had a devastating impact on native frog species. Of the 218 native species of frog described in Australia, 56 have been found to be infected with the chytrid fungus (DEH 2006b). It has caused the extinction of one species of frog and is suspected to have caused the extinction of three others (DEH 2006b). Evidence indicates that the amphibian chytrid fungus was introduced to southeast Queensland in the mid- to late-1970s (DEH 2006b), and subsequently has been recorded in four regions of Australia: east coast, Adelaide, southwest Western Australia (Berger 2001; Speare and Berger 2003a; Berger et al. 2004), and Tasmania (Obendorf 2005).

In NSW, 20 species, almost one-quarter of the total number of frog species occurring in NSW have been found to be infected with the disease (DEH 2006b). In August 2003, 'Infection of frogs by amphibian chytrid causing the disease chytridiomycosis' was listed as a Key Threatening Process (KTP) under Schedule 3 of the NSW *Threatened Species Conservation Act 1995* (TSC Act) (DECC 2003). Comparably, 'Infection of amphibians with chytrid fungus resulting in chytridiomycosis' was listed as a KTP under the Commonwealth *Environmental Protection and Biodiversity Conservation Act 1999* (EPBC Act) in July 2002 (DEH 2002).

The Department of Environment, Climate Change and Water NSW (DECCW) has identified various strategies and priority actions relevant to this threat in NSW, through the NSW Priorities Action Statement (PAS). A threat-based approach enables DECCW, where possible, to prevent other species from becoming threatened and is an efficient tool for addressing a threat that may affect a number of species and their habitat. A cooperative effort is required to mitigate the impact and manage the threat of infection by amphibian chytrid fungus by engaging with landholders, researchers and the public to prevent the further spread of the pathogen into uninfected areas and frog populations in NSW.

This Statement of Intent outlines DECCW's approach to implementing a broad program of actions over the next five years to manage the threat and minimise the spread and impact of chytridiomycosis on the frog species of NSW. Priority initiatives include:

- actions to address any gaps in knowledge
- promotion of effective hygiene practices to control the spread of the pathogen
- assessment of the risk posed to threatened species and populations of frogs in NSW and management of the threat at key locations
- raising of awareness through training, education and communication
- delivery of a coordinated threat abatement program.

Threat abatement actions contained in this Statement of Intent are consistent with those presented in the 'National Threat Abatement Plan: Infection of amphibians with chytrid fungus resulting in chytridiomycosis'. DECCW initiatives will be developed and implemented in conjunction with the national approach. DECCW envisages that this document will provide opportunities to work in partnership with other government agencies, industry, and land managers, and integrate efforts to tackle this threat into the future.

What is a Statement of Intent?

A Statement of Intent presents a strategic framework for a targeted five-year period threat abatement program relevant to DECCW's area of responsibility. It responds to priority actions identified in the 'National Threat Abatement Plan: Infection of amphibians with chytrid fungus resulting in chytridiomycosis' and implements actions from the NSW PAS.

Threat abatement planning is all about cooperation. Although this Statement of Intent is focussed on DECCW's area of responsibility, successful threat abatement relies on an integrated and collaborative approach involving other public authorities, organisations and the community. This Statement of Intent aims to promote coordination by detailing how DECCW is working in partnership to abate the threat from chytridiomycosis.

A Statement of Intent is similar to a Threat Abatement Plan (TAP) in that it details a threat abatement strategy for a listed Key Threatening Process; however, it is not a statutory document. A NSW TAP is a statutory document prepared and approved in accordance with Part 5 of the TSC Act, and Ministers and public authorities are required to take any appropriate action available to them to implement the measures in the plan. A Commonwealth TAP has already been prepared under Division 5 of the EPBC Act to help abate the threat from chytridiomycosis. The NSW Government collaborates with the Australian Government to implement any National TAP prepared under the EPBC Act and DECCW facilitates cooperation across the key government agencies and other stakeholders in NSW.

2. Distribution

Chytridiomycosis has been recorded from Australia, New Zealand, Europe, Africa, Asia and South, Central and North America from a broad range of amphibian habitats (Berger et al. 1999; Mutschmann et al. 2000; Waldman et al. 2000; Bosch et al. 2001; Fellers et al. 2001; Waldman et al. 2001; Bradley et al. 2002; Bonaccorso et al. 2003; Kusrini et al. 2008). The earliest recorded case of amphibian chytrid fungus infection was from the African clawed frog (*Xenopus laevis*) in South Africa in 1938, and epidemiological data supports that *Batrachochytrium dendrobatidis* originated in Africa (Weldon et al. 2004).

In Australia, chytridiomycosis has been found in frogs from four geographic areas: a large eastern coast zone extending from Big Tableland near Cooktown in the north to Melbourne in the south, an Adelaide zone, a southwest zone which includes the whole of south-west Western Australia to just north of Perth (Berger 2001; Speare and Berger 2003a; Berger et al. 2004), and Tasmania (Obendorf 2005).

In NSW and other parts of eastern Australia, the majority of reported chytridiomycosis cases have occurred between the Great Dividing Range and the coast, with high altitude (>400m) populations appearing to be more severely affected (Speare 2001); map 1 shows where ill or dead, free-living amphibians have been found infected with *B. dendrobatidis* in NSW.

The sensitivity of *B. dendrobatidis* to temperature appears to limit its spread, preventing it from becoming established or affecting frogs in locations where temperatures are consistently high (DEH 2006b). However, once frogs in a location are found to be infected, *B. dendrobatidis* appears to persist in that location irrespective of the density of the amphibian population (DEH 2006b).

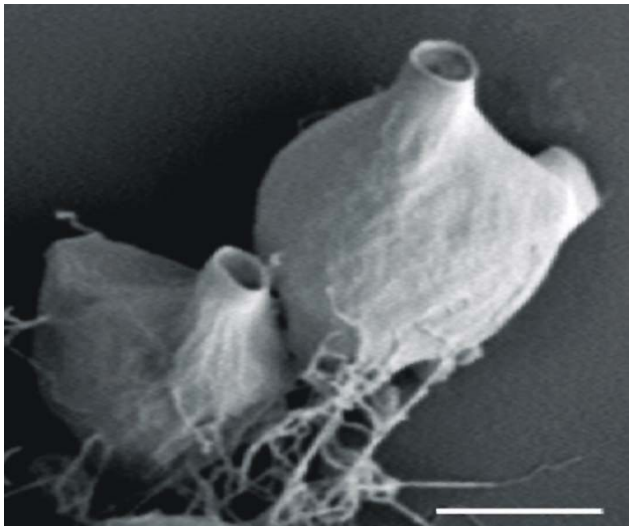
Map of NSW showing where ill or dead, free-living amphibians have been found infected with *B. dendrobatidis*.

1. Mebbin	2. Numinbah Valley	3. Boomi Creek	4. Nightcap Range	5. Peacock Creek
6. Tooloom Range	7. Yabbra	8. Lismore	9. Casino	10. Alstonville
11. The Basin	12. Mt Warning	13. Mann River	14. Grafton	15. Wild Cattle Creek
16. Lanitza	17. Bruxner Park	18. Boambee	19. Bonville	20. Dorrigo
21. Bellingen	22. Kalang River	23. Bowraville	24. Ellenborough River	25. Tapin Tops
26. Barrington Tops	27. Gloucester Tops	28. Muswellbrook	29. Chichester	30. Myall River
31. North Washpool	32. Maitland	33. Kooragang Island	34. Yengo National Park	35. Watagans Mountains
36. Davistown	37. Avoca	38. Sofala	39. Glenbrook	40. Springwood
41. Chester Hill	42. Homebush	43. Kurnell	44. Hacking River	45. Port Kembla
46. Kangaroo Valley	47. Hoskinstown	48. Budawang	49. Currawan Creek	50. Kosciuszko National Park
51. Mumbulla Falls	52. Broadwater Picnic Area			

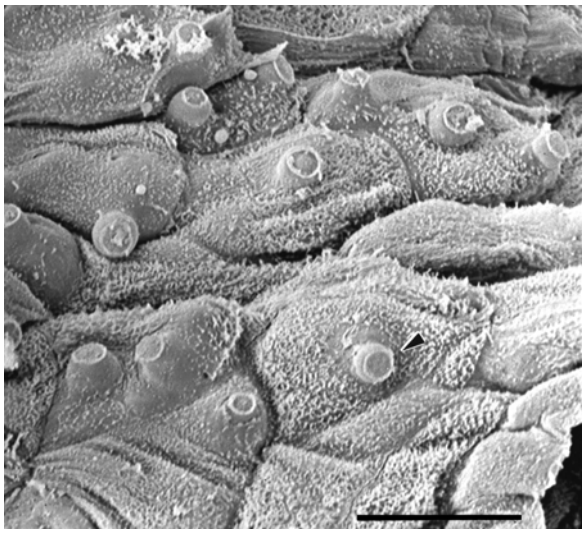
Source: Berger et al. 2004; Kriger et al. 2007; Mahony M, 2009, pers. comm. (University of Newcastle).

3. Biology and ecology of *Batrachochytrium dendrobatidis*

Batrachochytrium dendrobatidis (Berger et al. 2004) is a fungus in the Phylum Chytridiomycota, Class Chytridiomycetes, Order Chytridiales (Longcore et al. 1999). *B. dendrobatidis* is the only chytrid fungus known to infect vertebrate species (DEH 2004). It reproduces asexually and appears in two main forms; a spherical sessile zoosporangium 10–40µm in diameter, and a motile flagellated zoospore about 2µm in diameter. Zoospores form within the zoosporangium and are released into the external environment via discharge tubes (Berger 2001). The motile zoospore attaches to the substrate, develops rhizoids, and becomes a zoosporangium.



Zoosporangia of *B. dendrobatidis* growing *in vitro*. Note the open discharge tubes through which the zoospores escape.
Scale bar = 10µm
Source: Berger (2001).



Surface of the epithelium of a green tree frog (*Litoria caerulea*) with chytridiomycosis showing discharge tubes of zoosporangia of *B. dendrobatidis* emerging from the surface (arrowheads).
Scale bar = 10µm
Source: Berger et al. (1998).

The zoospore is transmitted via water and then invades the skin of amphibians (Berger et al. 1998; Longcore et al. 1999; Pessier et al. 1999; Nichols et al. 2001). The zoosporangia only grow in the keratinised layers of the epithelium. The zoosporangia are most commonly found on the feet and ventral skin of the body and limbs. Discharge tubes penetrate through to the surface of the skin and allow zoospores to escape into the environment to continue the life cycle (DEH 2006b). They can remain motile for over 24 hours (Berger 2001).

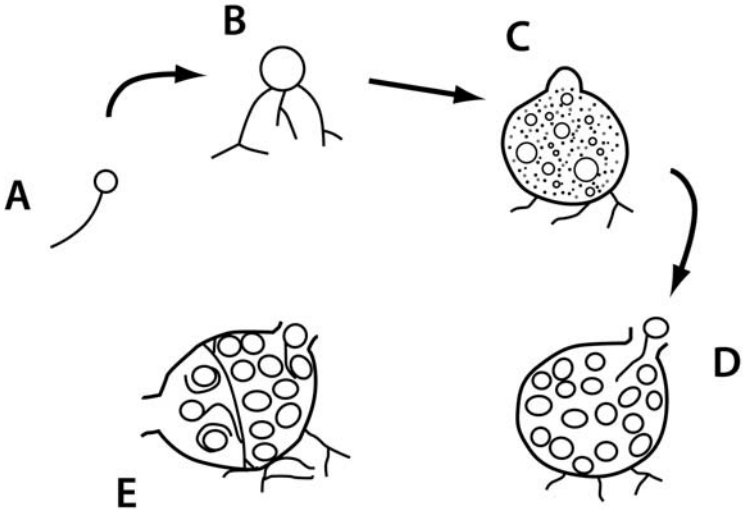


Diagram of the lifecycle of *B. dendrobatidis* in culture.
After a period of motility, zoospores (A) encyst, resorb their flagella and form germlings (B).
Rhizoids appear from one or more areas. Sporangia grow larger and mature over 4–5 days (C). The sporangia become multinucleate by mitotic divisions and the entire contents cleave into zoospores while the discharge tubes form. The zoospores are released through the discharge tubes (D). Some thalli develop colonially with thin septa dividing the contents into multiple sporangia each with their own discharge tube (E).

Source: Berger (2001) (Not to scale).

The zoospores of *B. dendrobatidis* are infective to both frogs and tadpoles (Berger et al. 1999; Nichols et al. 2001). Although chytrid fungus can be fatal in adult frogs, it does not appear to cause mortality in tadpoles and has not been found growing on eggs (Berger et al. 1998). In early tadpole stages, *B. dendrobatidis* is only found in the mouthparts as these are the only keratinised skin on the body. In later tadpole stages, as the tail and feet become keratinised and the mouthparts are lost, zoosporangia begin to infect these other sites (Berger 2001; Marantelli et al. 2004).

Experimental studies on susceptible species have shown that incubation times vary from 9 to 76 days, with death from chytridiomycosis generally occurring 18–48 days after exposure. However, infection is not fatal in all species of amphibians, and apparently healthy amphibians, such as bullfrogs (*Rana catesbeiana*), may frequently carry light infections (Mazzoni 2000; Mazzoni et al. 2003).

It is not yet known exactly how chytridiomycosis kills frogs. It is possible that the sporangia may release fatal toxins that are absorbed through the skin, or that the disease may directly affect the skin's osmoregularity, the ability to uptake water and respire through the skin (Berger et al. 1998; Pessier et al. 1999; Voyles et al. 2007).

Factors that may influence the expression of chytridiomycosis and have been shown to affect *B. dendrobatidis in vitro* include temperature, dehydration, salinity, water pH, light, nutrition and dissolved oxygen (Berger et al. 1999). Studies have shown that *B. dendrobatidis* grows slowly at 6°C and develops most rapidly at 23°C, but will not survive temperatures above 29°C (Longcore 2000; Berger 2001; Johnson et al. 2003). The influence of temperature on *B. dendrobatidis* may explain why populations found at high altitudes (>400m) are more severely affected by chytridiomycosis (Berger et al. 2004; McDonald et al. 2005).

B. dendrobatidis does not form a resting stage and does not survive drying; it therefore requires water or direct contact between frogs in order to be transmitted. It can spread slowly over the landscape by natural methods, but movement over long distances such as across oceans and deserts is likely to be due to human-assisted translocation of infected amphibians or water and soil contaminated with the pathogen (DEH 2006b).

Climate change and chytridiomycosis

The relationship between recent outbreaks of amphibian chytridiomycosis and climate change continues to be the subject of ongoing research (Lips et al. 2008; Bosch et al 2007; Skerratt et al. 2007; Laurance 2008; Rohr et al. 2008). Climate change represents a serious threat to biodiversity and is likely to influence the interaction between frogs and *B. dendrobatidis*. Just as temperature, rainfall, and humidity all influence the biology of frogs, these factors also affect the growth, persistence, and ecology of *B. dendrobatidis*.

Changes in regional or local climate may directly or indirectly alter the development, survival rates and transmission of *B. dendrobatidis* and host susceptibility, and thereby influence the host response (Lips et al. 2008). However, the results of studies conducted to date suggest that the recent outbreaks of chytridiomycosis are not linked to climate change.

4. Disease symptoms

The clinical signs of chytridiomycosis in Australian frogs manifest in three ways:

1. changes in behaviour, including lethargy, lack of appetite, sitting unprotected during the day, with hind legs spread slightly away from the body (Berger et al. 1999)
2. neurological signs, including reduced or loss of righting reflex (DEH 2006b)
3. skin lesions, including darkening and patchy discolouration of skin, sloughing or peeling on the outside layers of skin, erosions and ulcerations.

A great barred frog (*Mixophyes fasciolatus*) in the terminal phases of chytridiomycosis, with depressed attitude, half closed eyes and accumulation of sloughed skin over the body
Scale bar = 5 mm
Source: Berger (2001).

Although these clinical signs are indicative of the disease, they can in some cases be similar to those of other amphibian diseases, so laboratory testing is required for the diagnosis of chytridiomycosis (Berger et al. 1998; Berger and Speare 1998; Berger et al. 1999; Berger et al. 2000; Hyatt et al. 2000; Hyatt 2003). In addition, healthy frogs can also carry infections with no visible expression of the disease (DEH 2006b).

Innate resistance to *B. dendrobatidis* appears to be present in some individuals and in some species. As the abundance of frogs in some areas is increasing despite the presence of *B. dendrobatidis*, this suggests resistance may be evolving in these populations. However, the components of the immune system that are mobilised against *B. dendrobatidis* have yet to be determined (DEH 2006b).

Until relatively recently, direct examination of skin scrapings and histological examination of toe clip samples was the most widely used technique to diagnose chytridiomycosis (Berger et al. 2000, 2002; Berger 2001). However, another test known as the Taqman real time polymerase chain reaction (PCR) is now the preferred test for detection and considered to be the most accurate (Hyatt 2003; Boyle et al. 2004). PCR is a quantitative test which can detect one zoospore in a test sample and measure the total number of spores on a swab sample (Boyle et al. 2004). This information can be used to infer the level of infection on an individual frog. A major advantage of the PCR test is that it does not involve removal of a toe or destruction of the amphibian (DEH 2006b).

A section of skin from a heavily infected adult green tree frog (*Litoria caerulea*). Note immature stage (I), zoosporangium with discharge tube (D) containing zoospores, and empty zoosporangium after zoospores have discharged (arrow). E = epidermis.
Scale: largest zoosporangia are 15µm in diameter
Source: Berger et al. (2000)

5. Impacts on biodiversity

Frogs play an important role in the functioning of Australian ecosystems, and are an integral part of the food chain. Mature frogs eat insects, small fish, and other small aquatic and terrestrial animals, while tadpoles feed on algae and influence water quality. In turn they provide food for fish, some large insects, snakes, lizards, larger amphibians, birds, and small carnivorous and omnivorous mammals. Frogs are also considered good indicators of significant environmental changes as they are more vulnerable and sensitive to factors such as disease, pollution, toxic chemicals, radiation and habitat destruction than many other animals.

B. dendrobatidis is a highly virulent pathogen that is capable of infecting any species of amphibian; however, the intensity of infection and the pathological effects that *B. dendrobatidis* has on a population depends on a complex interaction between the specific characteristics of individual species, the pathogen and the environment, including how the species is distributed across a range of environments. Both innate and acquired immunity appears to be present in some individuals, populations and species. Therefore, the impacts on a species can range from extinction of the species as the worst case scenario, to a stable population where the fungus is present but sporadic deaths occur due to chytridiomycosis (DEH 2006b). The disease has been known to be active for at least 15 years in some south-west Western Australia species, such as the orange-bellied frog (*Geocrinia vitellina*) and the western green and golden bell frog (*Litoria moorei*), without an obvious impact on their populations (DEH 2004).

Across Australia

B. dendrobatidis has the potential to infect all native species of amphibian in Australia, although it does not prove fatal in all. Of the 28 species listed as threatened under the EPBC Act, chytridiomycosis is known to be active in 14 (DEH 2006b). It has also been found in an additional three native species in captivity, and in two introduced amphibians – the cane toad (*Bufo marinus*) (wild and captive populations) and the axolotl (*Ambystoma mexicanum*) (captive). Of the five amphibian families in the wild in Australia, chytridiomycosis has been reported in three: *Hylidae* (tree frogs), *Myobatrachidae* (ground frogs) and *Bufonidae* (toads; DEH 2006b).

At least one Australian species, the sharp-snouted day frog (*Taudactylus acutirostris*), has been driven to extinction by chytridiomycosis (DEH 2006b). Chytridiomycosis is also suspected to have caused the extinction of three other native species – the northern gastric-brooding frog (*Rheobatrachus silius*), the southern gastric-brooding frog (*Rheobatrachus vitellinus*), and the southern day frog (*Taudactylus diurnus*) (Berger 2001). Several examples are known where chytridiomycosis has also caused extinctions of localised populations of other species (DEH 2006b).

The Australian frog species threatened by chytridiomycosis have several characteristics in common. They are mostly from higher altitude areas, have significantly smaller clutch sizes, and occupy restricted geographic ranges (Williams and Hero 1998; McDonald and Alford 1999). Populations in the Cape York Peninsula, the Gulf Country in Queensland, northwest Western Australia, the Northern Territory and most of central Australia are currently thought to be chytridiomycosis-free (DEH 2006b).

Across New South Wales

Of the 84 native species of frog in NSW, chytridiomycosis has been reported in wild populations of 20 species (DEH 2006b). Of these, 13 are listed as threatened under the EPBC Act and 15 are listed as threatened under the TSC Act, as detailed in Table 1. Chytridiomycosis is also threatening the New England Tableland population of tusked frog (*Adelotus brevis*) which is listed as Endangered under the TSC Act.

Table 1 Chytridiomycosis infection has been reported in the following threatened frog species in NSW

Common name	Scientific name	EPBC Act	TSC Act
Green and golden bell frog	*Litoria aurea*	Vulnerable	Endangered
Booroolong frog	*Litoria booroolongensis*	Endangered	Endangered
Spotted tree frog	*Litoria spenceri*	Endangered	Endangered
Peppered frog	*Litoria piperata*	Vulnerable	Vulnerable
Glandular frog	*Litoria subglandulosa*	—	Vulnerable
Giant burrowing frog	*Heleioporus australiacus*	Vulnerable	Vulnerable
Stuttering frog	*Mixophyes balbus*	Vulnerable	Endangered
Fleay's barred frog	*Mixophyes fleayi*	Endangered	Endangered
Giant barred frog	*Mixophyes iteratus*	Endangered	Endangered
Red-crowned toadlet	*Pseudophryne australis*	—	Vulnerable
Southern corroboree frog	*Pseudophryne corroboree*	Endangered	Endangered
Yellow-spotted bell frog	*Litoria castanea*	Endangered	Endangered
Northern corroboree frog	*Pseudophryne pengilleyi*	Vulnerable	Vulnerable
Alpine tree frog	*Litoria verreauxii alpina*	Vulnerable	Endangered
Littlejohn's tree frog	*Litoria littlejohni*	Vulnerable	Vulnerable
Tusked frog population (Nandewar and New England Tablelands Bioregions)	*Adelotus brevis*	—	Endangered population

Source: NSW Scientific Committee (2003)

Chytridiomycosis also has the potential to cause a number of NSW frog species, which are currently not listed as threatened, to become threatened, as detailed in Table 2.

Table 2 NSW frog species which are currently not threatened, but may become threatened due to chytridiomycosis

Species name	Scientific name
Eastern banjo frog	*Limnodynastes dumerilii*
Blue Mountains tree frog	*Litoria citropa*
Lesueur's frog	*Litoria lesueuri*
Leaf-green tree frog	*Litoria phyllochroa*

Source: NSW Scientific Committee (2003)

Further surveys are required to identify additional species and populations in NSW that may be, or potentially become, threatened by chytridiomycosis. Targeted surveys have confirmed the presence of chytridiomycosis in key populations of some species however with only limited surveys in some areas, the full extent of distribution remains unknown.

Case study – Chytridiomycosis threatens the southern corroboree frog

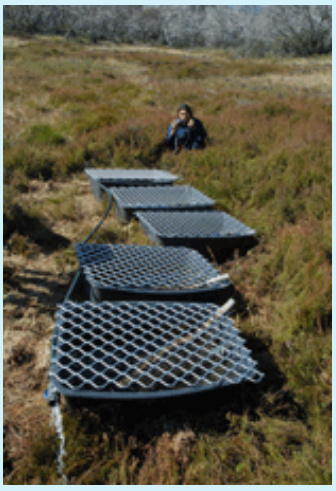
The southern corroboree frog (*Pseudophryne corroboree*) is listed as Endangered under both the TSC Act and the EPBC Act. Chytridiomycosis is a key threat to the survival of this species, with numbers declining to less than 200 breeding individuals in the wild over the past three decades (Hunter 2008). The southern corroboree frog is limited to high montane and sub-alpine bog habitats of the northern Snowy Mountains, with its range occurring entirely within Kosciusko National Park (Osborne et al. 1996). The Endangered listings triggered the implementation of threat abatement measures at the population sites, including:

- strengthening site hygiene protocols to prevent any transfer of infection
- establishing a captive breeding colony and reintroduction program
- monitoring the health of reintroduced individuals over time
- field research and experiments.

A number of animal husbandry institutions have been involved in the successful breeding and rearing of southern corroboree frogs in captivity; including the Amphibian Research Centre, Taronga Zoo, Melbourne Zoo, and Healesville Sanctuary. Field research conducted by DECCW has shown that reintroduced southern corroboree frog adults have bred successfully in the wild.

Research undertaken by DECCW has also implicated the common eastern froglet (*Crinia signifera*) as a reservoir host for *B. dendrobatidis* in southern corroboree frog breeding pools, and has demonstrated that reintroducing egg and tadpoles into artificial pools that cannot be accessed by common eastern froglets greatly reduces infection prior to metamorphosis (Hunter et al. 2007). It is hoped that measures will enable the southern corroboree frog to continue breeding in the wild and potentially develop a natural resistance against the disease.

Southern corroboree frog (*Pseudophryne corroboree*)



Artificial tadpole pools

Photos: David Hunter, DECCW.

6. What is being done nationally?

In 2006, the Australian Government developed the 'National Threat Abatement Plan: Infection of amphibians with chytrid fungus resulting in chytridiomycosis' in consultation with the states and territories. It aims to reduce the impact of chytridiomycosis on native frog species.

The National Threat Abatement Plan

The National Threat Abatement Plan focuses on strategic approaches to reduce the impact of chytridiomycosis to maximise the chances of the long-term survival of native species and ecological communities affected by the Key Threatening Process.

Under the EPBC Act, the Australian Government is responsible for implementing the plan in Commonwealth areas but also seeks the cooperation of the states and territories where the disease impacts within their jurisdictions. The Australian Government also supports the national effort through financial assistance for key national level actions in the plan, such as research and demonstration model projects that can be provided to landholders and managers to assist with effective threat management. The plan sets out an approach to achieving these goals by implementing currently available management strategies for control of chytridiomycosis, providing for the development of new techniques, and collecting information to improve our understanding of the extent and impacts of the pathogen in Australia (DEH 2006a).

This threat abatement plan has two broad goals:

- to prevent amphibian populations or regions that are currently chytridiomycosis-free from becoming infected by preventing further spread of *B. dendrobatidis* within Australia
- to decrease the impact of infection by *B. dendrobatidis* on populations that are currently infected.

The National Threat Abatement Plan and Background Document are available on the Australian Government's Department of the Environment, Water, Heritage and the Arts website at the following link:

http://www.environment.gov.au/biodiversity/threatened/publications/tap/chytrid.html

Research focusing on understanding the ecology of amphibian chytrid and its relationships with host species is being led by several institutions nationally, including:

- the Amphibian Diseases Research Group at James Cook University, Townsville, Queensland
- the Amphibian Research Centre, Werribee, Victoria
- the Animal Health Laboratory, Geelong, Victoria, within the Commonwealth Scientific and Industrial Research Organisation (CSIRO)
- the University of Newcastle, Callaghan, NSW.

A number of other institutions nationally have been involved in a variety of amphibian chytrid related research captive breeding programs. These include Taronga Zoo, the University of Queensland, the Queensland Parks and Wildlife Service, the University of Western Sydney, NSW Department of Primary Industries (Forests NSW), Arthur Rylah Institute, Department of Sustainability and Environment Victoria, The University of Tasmania, The University of Adelaide, the Western Australian Museum and The University of Western Australia.

7. What is being done in NSW?

DECCW is supporting a range of chytridiomycosis threat abatement work in NSW, including surveys to determine distribution and assess impacts, on-ground management, monitoring and research. DECCW has been implementing individual frog species conservation programs for a number of years.

A DECCW Amphibian Chytrid Threat Abatement Working Group was established to guide the planning and implementation of a state-wide threat abatement program for the pathogen, bringing together experts across the various DECCW divisions and groups. A NSW Declining Frog Working Group is currently operating to improve communication and information sharing between stakeholders (including DECCW, other government agencies, research institutions and private industry) that are involved in frog-related surveys, and conservation, research and monitoring projects within NSW. The group influences the direction of research and provides advice on government policy relating to the management of chytridiomycosis and its impacts within NSW.

DECCW National Parks and Wildlife Service has also developed the ‘Hygiene protocol for the control of disease in frogs’ (NPWS 2001). The hygiene protocol describes hygiene precautions to be undertaken when handling frogs and tadpoles to minimise the transfer of the pathogen.

Through undertaking these activities, DECCW has commenced implementing actions identified in the National TAP.

Chytrid threat abatement work undertaken by DECCW

Captive-breeding and reintroduction programs

Captive breeding and restocking key populations can give a threatened frog species a 'window of opportunity' to recover when faced with a mortality rate that will drive it to extinction. Restocking may buy time to allow the species to develop its own resistance or to survive a particularly hazardous situation due to a combination of adverse environmental, pathogen and host factors (McDonald et al. 2005).

DECCW is currently managing captive breeding and reintroduction programs for the following species threatened with extinction – spotted tree frog, southern corroboree frog, booroolong frog, and green and golden bell frog. DECCW is implementing programs to monitor the health of reintroduced individuals at these sites over time.

On-ground mitigation and site controls to protect threatened species

Chytridiomycosis is thought to have played a major role in the decline of the green and golden bell frog in the Sydney area and the southern corroboree frog in Kosciusko National Park. It has also been largely responsible for the failure of subsequent reintroduction efforts to recover these species.

DECCW has modified the habitat of these species at key sites in an attempt to improve survival rates of reintroduced captive bred frogs and tadpoles. Additional measures undertaken at these sites include strengthening site hygiene protocols and conducting field research and experiments into the interaction between the disease and the species (DECC 2008a, b, c).

Treatment of individuals

Effective treatments in adults and tadpoles are needed to prevent mortalities in captive programs for threatened species, and will reduce the risks associated with the movement of amphibians (DEH 2006b). A recent discovery by researchers at the University of Otago in New Zealand suggests that the antibiotic chloramphenicol may be capable of curing chytridiomycosis in individual frogs in captivity (Poulter et al. undated). In addition, a another study has shown that raising the temperature of infected frogs to 37°C resulted in the cure of asymptomatic chytridiomycosis in some species (Woodhams et al. 2003). Itraconazole has also been used to effectively treat chytridiomycosis in some species (Nichols et al. 2001).

Green and golden bell frog (*Litoria aurea*).
Photo: Dave Hunter. DECCW

8. Key challenges and initiatives

This section describes the key challenges and the responding initiatives DECCW will pursue as part of a targeted threat abatement program to tackle infection of frogs by amphibian chytrid causing the disease chytridiomycosis over the next five years. These actions are grouped under the five main challenges currently faced, in order to effectively control the infection and prevent the spread of the pathogen in NSW. DECCW's approach to managing this KTP will be dynamic as information from research into the impacts and potential management response to chytridiomycosis is presented.

The costs will be sourced from DECCW recurrent budgetary allocations and external grants. The indicative timeframes allocated to each action are identified as either:

(S) short term, i.e. 1–3 years

(M) medium term, i.e. 3–5 years

(L) long term, i.e. >5 years.

Challenge 1: Understanding the threat

Although significant progress has been made in recent years, further research is required to understand the ecology and impact of chytridiomycosis on NSW frogs. Investigations will be challenged by:

- the limited number of thorough and systematic surveys undertaken of species that have declined since the introduction of *B. dendrobatidis*
- the limited area of NSW that has been surveyed for infection status to date
- the cost of diagnostic testing of surveyed populations
- the need to establish site-based surveying and monitoring of long-term effects
- the potential effects that climate change may have on the distribution and virulence of the pathogen.

Initiative 1: Undertake research, surveying and monitoring of the pathogen

DECCW will:

1.1 Support research into transmission and dispersal of *B. dendrobatidis* to improve understanding of the disease through monitoring key threatened frog populations and trialling habitat modification techniques (S)

1.2 Conduct surveys of priority NSW locations thought to be free of chytridiomycosis (identified in the National TAP) to improve knowledge of the distribution of the pathogen and to identify wild populations most at risk (S)

1.3 Develop a system for reporting chytridiomycosis infection (both by DECCW and the public) and maintain a database of sites in NSW where the disease has been identified or is suspected of being present (S/M/L)

1.4 Support research into understanding species resistance to *B. dendrobatidis*, both innate and acquired, including the comparison of chytridiomycosis-free populations identified in action 1.2 with infected populations to assess evolutionary responses, and to potentially improve the success of reintroduction programs (S).

Cooperative chytridiomycosis research

DECCW is participating in two collaborative projects that address key research and monitoring actions 1.1 and 1.2 of this Statement of Intent.

The first project will be funded by the Australian Biosecurity Cooperative Research Centre for Emerging Infectious Disease and will be undertaken in conjunction with James Cook University, Taronga Conservation Society Australia, the University of Newcastle, the Amphibian Research Centre, Western Australia Department of Environment and Conservation, Tasmania Department of Primary Industries and Water, and the Australian Wildlife Health Network.

This project will involve:

- studying captive-bred booroolong frogs to investigate the possibility of inducing adaptive *B. dendrobatidis* immunity in frogs (in this case, using chloramphenicol), to improve the success of threatened species reintroduction programs
- capturing alpine tree frogs from wild populations with varying histories of chytridiomycosis exposure to establish separate captive populations, and determine whether selection for innate immunity has occurred naturally and can potentially be used to improve reintroduction success when disease is the threatening process
- research to improve our understanding of how innate immunity to *B. dendrobatidis* in amphibians is selected for.

The second project will be funded by the Australian Research Council and will be undertaken in cooperation with Sydney Olympic Park Authority, the University of Newcastle, Strathfield Council, South Australian Museum and NSW Roads and Traffic Authority. The project integrates investigations in population ecology, dynamics and genetics, and disease ecology to secure the population of endangered green and golden bell frogs at Sydney Olympic Park. This investigation into the role of chytridiomycosis in the population decline, and the effect that habitat modification has had in reducing the impact of chytridiomycosis, will provide valuable information for future mitigation measures.

Challenge 2: Controlling the spread of the pathogen

As knowledge of the factors contributing to the spread of the pathogen improve, management efforts should also focus on precautionary measures to control any spread by humans by modifying behaviour and activities. This should include control of the spread of chytrid fungus both from infected to chytridiomycosis-free areas, but also between populations and individuals in chytrid-infected areas to minimise impacts from the disease (DEH 2006b). This includes monitoring and managing the movement of frogs during research, trade and those displaced through the movement of agricultural and other produce.

Initiative 2: Identify and promote effective hygiene practices to control the spread of the pathogen in NSW

DECCW will:

2.1 Update existing hygiene protocols and promote their use by researchers, ecologists, government agencies, park/zoo operators, wildlife rescue and carer organisations, frog interest groups and other individuals who regularly deal with or are likely to handle frogs (S)

2.2 Reduce the risk of pathogens spread by the movement of individuals, equipment or substrates within DECCW estate, by implementing hygiene protocols at both the potential source and destination of infection (S/M/L)

2.3 Mitigate the impact of the disease by implementing hygiene protocols in infested areas within DECCW estate (S/M/L).

Hygiene protocols to manage the spread of *B. dendrobatidis*

Controlling the human-facilitated movement of amphibians between sites and ensuring that appropriate hygiene protocols are followed by people handling frogs or substrates from frog habitats are essential in controlling the spread of *B. dendrobatidis*. Humans have the potential to spread the pathogen further and faster than any other vector, through the movement of infested soil, water, or infected frogs and tadpoles. The National TAP promotes best practice to control the spread of *B. dendrobatidis* across the landscape by modifying human behaviour (DEH 2006b).

DECCW National Parks and Wildlife Service developed the 'Hygiene protocol for the control of disease in frogs' (NPWS 2001). The hygiene protocol describes hygiene precautions to be undertaken when handling frogs and tadpoles to minimise the transfer of the pathogen on footwear, survey equipment and vehicles. It outlines measures to:

- prevent or reduce *B. dendrobatidis* being transferred within and between wild populations of frogs
- ensure captive frogs are not infected prior to release
- deal safely with unintentionally transported frogs
- assist with the proper identification and management of sick and dead frogs in the wild (NPWS 2001).

Effective disinfection protocols are essential to ensure that human actions, particularly amphibian research, do not contribute to the spread of this pathogen. The hygiene protocol is intended for use by researchers, ecologists, government agencies, park/zoo operators, wildlife rescue and carer organisations, frog interest groups and other individuals who regularly deal with or are likely to handle frogs (NPWS 2001), or undertake work within frog habitats.

The protocol also contains procedures for the management of displaced frogs, commonly known as 'banana box frogs', that are often accidentally moved in agricultural produce. It also provides advice in dealing with sick or dying wild frogs infected with the amphibian chytrid.

The 'Hygiene protocol for the control of disease in frogs' can be found on the DECCW website under the following link:
http://www.environment.nsw.gov.au/resources/nature/hyprfrog.pdf

Challenge 3: Identifying and implementing effective mitigation measures

At present, there are no known methods for eradicating *B. dendrobatidis* once it has become established in a natural ecosystem, and there are currently limited methods available to control the pathogen at infected sites. The actions under this initiative seek to improve DECCW's ability to effectively manage populations in which *B. dendrobatidis* is present in order to minimise further declines. In order to inform future management decisions, there is an urgent need to monitor the effectiveness and efficiency of threat abatement measures and recovery strategies.

Initiative 3: Manage the threat of chytridiomycosis posed to threatened species and populations of frogs at key locations

DECCW will:

3.1 Undertake risk assessment of NSW listed threatened frog species and populations to identify and prioritise those most at risk from chytridiomycosis (S)

3.2 Implement NSW PAS actions for threatened species, threatened populations and the KTP, in order of priority identified in action 3.1, to minimise the threat from chytridiomycosis (S/M/L)

These may include:

- emergency responses
- habitat modification
- captive breeding programs, selective breeding for resistance and reintroductions
- treatment of individuals with antifungal chemicals such as chloramphenicol and induction of acquired immunity through exposure and treatment
- translocation of individuals/populations.

NSW PAS actions for chytrid can be viewed at: http://www.threatenedspecies.environment.nsw.gov.au/tsprofile/pas_ktp_profile.aspx?id=20009

3.3 Monitor priority population sites to determine the effectiveness of management strategies, and to determine the long-term impact of the pathogen (S/M/L).

Challenge 4: Improving awareness and capacity to manage the threat

There is a need to effectively communicate the actual and potential impacts of chytridiomycosis to stakeholders such as land managers (including public authorities), consent and determining authorities, ecologists, government agencies, bushwalkers, primary producers and those involved in the amphibian trade. Several government agencies and industry groups play a key role in the capacity to minimise the spread of chytrid fungus in NSW. The actions under this initiative seek to raise awareness of chytridiomycosis among stakeholders and to improve their capacity to manage the threat effectively.

Initiative 4: Strengthen training, education and communication

DECCW will:

4.1 Establish and/or maintain networks and share information with the Australian Government, state agencies, catchment management authorities (CMAs), local government areas and research organisations in relation to the management of the pathogen (S/M/L)

4.2 In partnership with other state agencies, develop and distribute community education and/or awareness-raising materials on chytridiomycosis to individual stakeholder groups including the agricultural sector (S/M)

4.3 Establish demonstration sites to promote the adoption of effective management techniques (S/M).

Challenge 5: Effectively coordinating the threat abatement program

Managing the threat of amphibian chytrid fungus across NSW is a complicated task due to the range of organisations that need to be engaged, the complexity of the threat, the extent of management actions, and the limited resources available. A coordinated approach will improve the effectiveness and ensure the timely, cost-effective and efficient delivery of a NSW threat abatement program.

Initiative 5: Effectively and efficiently implement an amphibian chytrid fungus threat abatement program

DECCW will:

5.1 Support the experts of the Amphibian Chytrid Threat Abatement Working Group to coordinate the NSW threat abatement program for this pathogen, consistent with this Statement of Intent and the National TAP, including responsibility for:

- 5.1.1 overseeing and monitoring the implementation of actions (as presented in this Statement of Intent) (Annual)
- 5.1.2 evaluating the effectiveness of existing resource effort and DECCW's capacity to manage the threat (resource effort to include measures of in-kind contribution of time, staff and re-current funding and external grants) (Annual)
- 5.1.3 investigating funding opportunities as required (S/M/L)
- 5.1.4 evaluating the uptake of relevant threat abatement actions, guidelines, protocols and priority assessment procedures across DECCW and externally
- 5.1.5 establishing partnerships with other government agencies, CMAs, research organisations and industry to integrate efforts to manage this threat in NSW.

5.2 Support the NSW Declining Frog Working Group in its role as a communication forum between experts and stakeholders from government agencies, research institutions and private industry (S/M/L)

5.3 Participate in the coordination of implementing the National TAP (S/M/L)

5.4 Develop, implement and report on actions in the PAS (S/M/L)

5.5 Coordinate incorporation of Statement of Intent actions into recovery plans and regional plans, such as Catchment Action Plans and Biodiversity Management Plans (S/M)

9. Where can I go for further information?

DECCW is coordinating delivery of actions presented in this Statement of Intent through the Amphibian Chytrid Threat Abatement Working Group.

For further information about the NSW Amphibian Chytrid Fungus threat abatement program or any other related projects, contact the DECCW Conservation Policy and Strategy Section, Landscapes and Ecosystems Conservation Branch on:

Tel: 9995 5000

Email: info@environment.nsw.gov.au

Useful web sites

- DECCW's NSW threatened species information: http://www.threatenedspecies.environment.nsw.gov.au/tsprofile/index.aspx
- Amphibian Diseases Research Group, James Cook University: http://www.jcu.edu.au/school/phtm/PHTM/frogs/ampdis.htm
- Amphibian Research Centre http://www.frogs.org.au

Further information is also available from the Australian Government's Department of the Environment, Water, Heritage and the Arts invasive species website at: http://www.environment.gov.au/biodiversity/invasive/index.html

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