Pathogenic skin fungi in Australian reptiles

Fact Sheet

September 2024

Key points

- Fungi from the genera *Nannizziopsis*, *Paranannizziopsis* and *Ophidiomyces* are the cause of skin diseases that may progress to systemic and sometimes fatal disease in a range of reptile species.
- These fungi were formerly members of the *Chrysosporium* anamorph of *Nannizziopsis vriesii* [CANV] complex, also referred to as 'yellow fungus disease'.
- These diseases are relatively newly described, suggesting they are 'emerging', although much remains to be learnt about these fungal diseases worldwide.
- It is not known if *N. barbatae* was introduced into Australia or if it is an endemic fungus however it is likely that global human-assisted movement of reptiles (due to the reptile pet trade) is contributing to the apparent emergence of these infections in both free-living and captive reptiles [1]. Altered host susceptibility or changing environmental conditions may also be contributing to disease emergence.
- In Australia, pathogenic skin fungi have been reported in wild lizards and fresh-water turtles, as well as in captive lizards, snakes and crocodiles.
- People working with captive and free-living reptiles should practice good biosecurity, as these pathogens are easily spread through contact.

The focus of this Fact Sheet is on fungi of the genera *Nannizziopsis*, *Paranannizziopsis* and *Ophidiomyces*.

Aetiology

The genera *Nannizziopsis* and *Paranannizziopsis* fall in the family *Nannizziopsidaceae* of the order Onygenales* [2]. *Ophidiomyces* falls in the family *Onygenaceae* (Onygenales) [3].

At least nine species of the genus *Nannizziopsis* are associated with skin disease in lizards globally [3-5]. *Nannizziopsis barbatae*† has 99% nucleotide similarity to *N. crocodili* and is also similar genetically to *N. pluriseptata* [5].

Genus *Paranannizziopsis* (four species) is linked to skin disease in Australasian reptiles [4, 6]. *Ophidiomyces ophidiicola*‡ (formerly *O. ophiodiicola*; the only species of this genus) is associated

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* The fungal agents formerly known as the *Chrysosporium* anamorph of *Nannizziopsis vriesii vriessi* (CANV complex) have been reclassified within the genera *Nannizziopsis*, *Paranannizziopsis* and *Ophidiomyces* (Sigler et al. 2013; Stchigel et al. 2013).

† Formerly *N. barbata*

‡ Formerly *O. ophiodiicola*

with “snake fungal disease” in terrestrial or semiaquatic snake species [2, 3]. Other onygenalean fungi include *Emydomyces testavorans* (turtles; no assigned family) [7] and *Aphanoascella galapagosensis* (land tortoise) [8].

One Health implications

Wildlife and the environment: *Nannizziopsis* spp. are increasingly being found in wild reptiles. They have the potential to cause significant disease and welfare issues in individual reptiles, and may exert a population-level impact. It is likely that global human-assisted movement of reptiles (due to the reptile pet trade) is contributing to the apparent emergence of these infections in both free-living and captive reptiles. While the list of affected reptile species continues to grow, there is no indication that wildlife other than reptiles are affected.

Domestic animals and humans: there is no evidence that pathogenic reptilian skin fungi will infect or cause disease in warm-blooded animals. Cases of *Nannizziopsis* infection in humans are caused by species distinct from those found in reptiles. The risk of zoonotic transmission of pathogenic skin fungi from reptiles to humans is considered low, since the temperature range for growth of these reptile associated fungi is generally not compatible with infection in humans [5].

Natural hosts

***Nannizziopsis* spp.** have been reported from infections in a wide range of lizard species (including chameleons, geckos, dragon and iguana lizards), fresh-water turtles and crocodiles [4, 5]. *Nannizziopsis guarroi* is associated with skin disease in bearded dragons (*Pogona vitticeps*) and other lizards in North America [4]. *Nannizziopsis barbatae* is increasingly linked to skin and systemic disease in a growing range of lizards in Australia (below), including endemic free-living species [5].

***Paranannizziopsis* spp**. have been reported in lizards, snakes and tuatara (*Sphenodon punctatus*) [4].

Ophidiomyces ophidiicola has been identified in a wide range of both captive and free-ranging snakes [2-4, 9].

Emydomyces testavorans has been associated with invasive shell and skin lesions in captive and free-living turtles in North America [7]. ***Aphanoascella galapagosensis*** has been isolated from a captive Galapagos tortoise with shell lesions in the USA [8].

World distribution and occurrences in Australia

Cases *of Nannizziopsis* spp. and *Paranannizziopsis* spp. infection in captive reptiles have been reported in Africa, Asia, Europe, North America, Australia and New Zealand [2-5]. Lesions typical of those associated with *O. ophidiicola* have been reported in captive snakes since the 1980s, primarily in North America but also in Europe. Infection has been identified more recently in wild snakes in North America [1-4, 9, 10].

Fungal species identified in Australia include *N. barbatae*, *N. crocodili*, *N. pluriseptata*, a *N. vriesii*-like fungus, *P. australasiensis* and *O. ophidiicola* [3-5, 11, 12] (see Table 1) .

Table 1: Onygenalean fungi associated with disease in Australian reptiles

Fungal species	Host species	Disease	Location	Captive/ wild	References
Nannizziopsis barbatae	Eastern water dragon (*Intellagama lesueurii*)	Dermatitis, debility, death	Brisbane, Qld	Wild	Peterson et al. 2020 [5]
N. barbatae	Tommy roundhead (*Diporiphora australias*)	Dermatitis, debility, death	Brisbane	Wild	Peterson et al. 2020 [5]
N. barbatae	Eastern bluetongue skink (*Tiliqua scincoides scincoides*)	Dermatitis, debility, death	Dubbo NSW	Wild	Peterson et al. 2020 [5]
N. barbatae	Shingleback lizard (*T. rugosa*)	Dermatitis, debility, death	Perth WA	Wild	Peterson et al. 2020 [5]
N. barbatae	Centralian bluetongue skink (*T. multifasciata*)	Dermatitis, debility, death	Vic	Captive	Peterson et al. 2020 [5]
N. barbatae	Coastal bearded dragon (*P. barbata*)[§]	Dermatitis	NSW	Captive	Paré and Sigler 2016 [4], Johnson et al. 2011 [11]
Nannizziopsis sp.	Coastal bearded dragon	Severe dermatitis, lost digits	SE Qld	Wild	eWHIS
N. barbatae	Macquarie turtle (*Emydura macquarii*)	Dermatitis	Sydney NSW	Wild	WHA 2021 [13] eWHIS
N. barbatae	Eastern snake-necked turtle (*Chelodina longicollis*)	Dermatitis	Sydney NSW	Wild	WHA 2021 [13] eWHIS
N. barbatae	Land mullet (*Egernia major*)	Dermatitis, death	Sydney NSW	Captive	eWHIS
N. barbatae-like	Shingleback lizard	Dermatitis	WA	Captive (wild-caught)	WA Wildlife Health Reference Group 2019 [14]
N. barbatae-like	Eastern water dragon (*Intellagama lesueurii*)	Dermatitis		Wild	Paré and Sigler 2016 [4]
N. vriesii-like	Rottnest Island shingleback (*T. rugosa konowi*)	Dermatitis	Perth WA	Wild	eWHIS
N. pluriseptata-like	Eastern Pilbara spiny-tailed skink (*E. epsisolus*)	Dermatitis, death	Sydney NSW	Captive	eWHIS
N. crocodili	Saltwater crocodile (*Crocodylus porosus*)	Dermatitis, death	Northern Qld	Captive (farmed)	Paré and Sigler 2016 [4], Thomas et al. 2002 [12]
N. crocodili	Freshwater crocodile (*C. johnstoni*) juveniles	Dermatitis		Captive (zoo)	Hill et al. 2019 [15]
Ophidiomyces ophidiicola	File snake	Dermatitis	Qld	Captive	Sigler et al. 2013 [3]
O. ophidiicola	Broad-headed snake (*Hoplocephalus bungaroides*)	Death	SA	Captive (zoo)	McLelland et al. 2010 [16]
Paranannizziopsis australasiensis	File snake (*Acrochordus arafurae*)	Dermatitis	Vic	Captive (zoo)	Sigler et al. 2013 [3], Paré and Sigler 2016 [4], Paré and Jacobson 2007 [17]
Paranannizziopsis spp. (suspect)	Coastal bearded dragon	Dermatitis	Qld	Wild	eWHIS

[§] Diagnosed as CANV at the time

Nannizziopsis spp. have increasingly been linked to disease in a range of wild (and captive) reptiles in Australia [5]. *Nannizziopsis crocodili* has only been reported in captive crocodiles and *Ophidiomyces ophidiicola* has been diagnosed only in captive snakes in Australia [3, 16].

***Nannizziopsis*:** the first global report of infection with this genus of fungi in wild reptiles were in four species of lizard across at least 8 different locations in Australia, including a significant outbreak among Eastern water dragons (*Intellagama lesueurii*) in Brisbane, Qld, beginning in 2013. *N. barbatae* caused severe, proliferative dermatitis, debility and death.

An outbreak in a group of captive coastal bearded dragons (*P. barbata*) in NSW in 2008-2009 (diagnosed as CANV at the time) was subsequently recognised [4, 11] as the first known cases of *N. barbatae* in Australia [3, 5, 11]. *Nannizziopsis barbatae* has not been reported outside Australia [1].

Anecdotal evidence suggests that clinical cases consistent with *Nannizziopsis* and *Ophidiomyces* fungal disease may have been occurring in captive lizards and snakes in Australia for several decades [18], although Peterson et al. 2020 [5] reports no positive cases from samples archived from 2010 onwards. It has been suggested that mycotic dermatitis historically attributed to *Geotrichum candidum* in three captive carpet pythons (*Morelia spilotes variegate*) in Qld may have been caused by *O. ophidiicola* [4, 19, 20].

Epidemiology

The epidemiology of these emerging diseases in reptiles is still poorly understood. Ectothermic taxa such as reptiles are considered particularly vulnerable to fungal pathogens. There is increasing evidence that at least some of these fungi (e.g. *N. barbatae*) are obligate pathogens [4, 5], rather than opportunists (as fungal pathogens of reptiles have been traditionally considered) [5, 21].

Nannizziopsis* and *Paranannizziopsis: Koch's postulates have been fulfilled in experimental infection of veiled chameleons (*Chamaeleo calyptratus*) with *N. guarroi* [22]. In captivity, suboptimal environmental factors may make reptiles more susceptible to these infections and the pathogens are contagious between animals in close proximity [4, 12]. There is little available information on the epidemiology in the wild [4, 5].

It is not known if *N. barbatae* was introduced to wild lizards in Australia through spillover or if it is an endemic fungus that has only been detected recently. It is not known if it is truly an "emerging disease" among wild lizards, nor what the factors might be that could be contributing to emergence, however altered host susceptibility or changing environmental conditions have been suggested as possible contributing factors [5].

Ophidiomyces ophidiicola is now well recognised as a pathogen of snakes, including free-ranging species particularly in North America. Experimental infections have fulfilled Koch's postulates and have demonstrated action as a primary pathogen [23, 24]. There is some evidence that some asymptomatic carrier hosts may exist, but are rare [1]. It is possible that the fungus can be vertically transmitted from dam to offspring (around the time of birth) [25] but transmission routes in general are poorly understood. The fungus is found in copious amounts in the lesions of affected snakes, and is not thought to be part of the cutaneous mycobiota [4, 26, 27]. Although the fungus does not grow at temperatures less than 15 C, snakes undergoing brumation, or held at suboptimal

temperature, may be susceptible if the snakes' immune systems are depressed [4, 9]. Small skin lesions that have been recognised for many years in snakes, colloquially termed "hibernation sores" or blisters, also test positive for the fungus, suggesting infection has been occurring historically, but the severity of clinical disease (at least in some incidences) may have been increasing in recent years. The cause of this increased severity (from small healing sores to a fatal disease) is unclear [4, 9]. Infection may make snakes more susceptible to predation or other causes of mortality and has been implicated as a cause of wild snake population declines since 2006 [10, 28].

Clinical signs

Lesions are similar for all pathogens, varying in severity depending on the host species and likely other environmental factors [5]. Grossly, lesions are initially yellow, and then thicken to form brown, hyperkeratotic, necrotic plaques. These plaques may crack and seep exudate, or slough to reveal whitish pink, swollen dermis. Lesions are often found on the head and around the mouth but can occur anywhere on the body and can involve a whole limb in the case of lizards [4, 5, 9, 17]. Animals may show hyperkeratosis, epidermal hyperplasia, dermal inflammation, necrosis, ulceration, and emaciation.

Figure 1. *Nannizziopsis* fungal disease in a captive coastal bearded dragon, *P. barbata* (Courtesy R Johnson).

Dermatomycoses due to *Nannizziopsis* spp. are slowly progressive and often fatal. Affected reptiles range in body condition from poor to good. Skin lesions progress over several months from dry and yellow to hyperkeratotic plaques to exudative and necrotic ulcers (Figure 1) [4, 5, 11]. In bearded dragons (*Pogona* spp.), the mouth and face are commonly affected but lesions may occur anywhere on the body [11, 29-32]. Infection is often fatal in bearded dragons, with infection extending to muscle, bone and internal tissues including liver, heart, kidney, lungs and intestine [4, 6, 11, 30, 32]. In shingleback lizards, infection may be mild and resolve with topical and/ or supportive treatment [33]. In iguanas, infection is often identified in the hind limbs and tail [34, 35]. Hatchling saltwater crocodiles infected with *N. crocodili* developed multiple leathery plaque-like lesions on or under the scales, which could be peeled away to reveal white or red tissue. Infection was often fatal [12].

Paranannizziopsis australasiensis infection In file snakes resulted in disseminated punctate or circular, whitish lesions across the epidermis [3, 4, 17].

Ophidiomyces ophidiicola in snakes mainly affects the head and also the scales of the body, most often on the underside of the snake. In milder cases, the processes of ecdysis (slough) can resolve the infection, but if deeper tissues are affected, disease may recur post-slough. Systemic infection with involvement of bone and lung has been reported, but is rare [9].

Diagnosis

Definitive diagnosis of skin or systemic disease associated with fungi in the *Nannizziopsis*, *Paranannizziopsis* and *Ophidiomyces* genera requires both a) identification of the organism through culture and sequencing or PCR and b) histopathology identifying fungal elements within lesions, in particular the presence of arthroconidia. Cytology of sticky tape preparations or impression smears, although not pathognomonic, may reveal conidia or arthroconidia suggestive of a fungus in the *Nannizziopsis*, *Paranannizziopsis* or *Ophidiomyces* genera [4, 17, 31]. Conidia are 5-8 µm by 3-5 µm, clavate to ovoid to cylindrical. Arthroconidia may be arranged in rows of up to 7 conidia, separated or in a chain [20, 31]. Presence of *Nannizziopsis*, *Paranannizziopsis* or *Ophidiomyces* is indicative of disease where hyphae and arthroconidia are present in lesions and the fungus has been isolated or confirmed by PCR. Identification of the organism using matrix-assisted laser desorption/ ionization time-of-flight mass spectrometry (MALDI-TOF MS), which may be more accurate than sequencing, has been described but may not yet be available in Australia [36]. A *N. barbatae*-specific PCR has been developed in Australia [37].

Laboratory diagnostic specimens

Multiple skin biopsies of dermal lesions, in 10% formalin (for histopathology) and submitted fresh (or less desirably frozen) for PCR and /or culture [4]. The fungus may be difficult to culture from swabs [4], however a *N. barbatae*-specific PCR showed high sensitivity when moderate or severe skin lesions where swabbed [37]. Sections of multiple internal organs in 10% formalin and fresh/frozen are recommended if systemic disease is suspected [38].

Pathology

Grossly, lesions are as described above. In systemically affected bearded dragons at necropsy, collection of pale yellow, gelatinous material can be found within the coelomic cavity and pericardial sac and granulomatous changes have been noted in the liver [30]. Histologic lesions include granulomatous fungal dermatitis, myositis, osteomyelitis, hepatitis, nephritis, coelomitis, myocarditis and pneumonia [4, 11, 30, 32].

Differential diagnoses

Other dermatomycoses, bacterial dermatitis, stomatitis and osteomyelitis should be excluded from the list of differential diagnoses.

Laboratory procedures

Histopathological examination should include H & E and either PAS or Grocott-Gomori's methylene silver stain for fungal identification [38]. Samples for culture are best treated with enrofloxacin to limit bacterial overgrowth, plated on Mycosel™ agar (Becton, Dickinson and Company, Franklin

Lakes, NJ) and incubated at 30°C. White powdery colonies should be sub-cultured, and speciated by sequencing [4]. Peterson et al. 2020 [5] recommends an improved method of fungal isolation by modification of the conventional serial dilution technique.

Treatment

Medical treatment of confirmed cases involves systemic antifungals and topical antifungal or antiseptic solutions [4]. Surgical excision or debridement of lesions may be carried out in conjunction with medical therapy [5]. There is limited information on successful medication regimens, and treatment may not be effective or curative [5]. Susceptibility testing of *N. guarroi* has revealed sensitivity to voraconazole and terbinafine, but less so to itraconazole [4, 39]. Pharmacokinetic studies in lizards suggests terbinafine may reach therapeutic levels after oral administration [40] and an infected bearded dragon was cured after prolonged treatment with systemic terbinafine [41]. Serum biochemistry should be monitored for signs of liver toxicity during antifungal treatment.

Treatment of captive reptiles should include appropriate cleaning and decontamination of the animal's environment, and the maintenance of simple surroundings to facilitate ongoing hygiene and disinfection. There is currently little information on appropriate disinfections protocols for *Nannizziopsis* sp. however laboratory-based studies on *O. ophidiicola* recommend a minimum of 2 minute environmental exposure to at least 3% bleach or 70% ethanol or a 10 min exposure to 0.16% Roccal, Lysol products, CLR, NPD, or 409 [42].

Prevention and control

Prevention of pathogenic fungal infection in captive reptiles should focus on reducing the fungal load, with attention being paid to regular substrate changes and good hygiene in captive situations. In captive reptiles, infection appears to be more common at ambient temperatures, below, or in the lower regions of the animal's preferred temperature range. Providing optimal husbandry conditions, including species-appropriate temperature gradients, hydration/humidity and nutrition are important steps in prevention and control. Affected individuals should be isolated and biosecurity measures followed as the organism can act as a contagious, primary pathogen [4].

Prevention and control options for free-living reptiles are limited. Factors that may contribute to increased susceptibility to infection such as environmental degradation, proximity to humans and other stressors should be addressed wherever possible [5]. Appropriate biosecurity and disinfection protocols (see above) should be used for equipment if working with wild reptiles, prior to moving between study sites [42].

Research

Molecular differentiation of the taxonomy of this order of fungi has significantly advanced knowledge in this area, allowing identification of morphological and physiological properties, host trends and sensitivity patterns for many new species [4]. There are significant knowledge gaps related to the incidence, host range and epidemiology of disease related to these pathogens, particularly in free-ranging reptiles [5, 9]. Further studies are warranted to understand fully the origin

and nature of these organisms in the Australian context, and their significance as primary pathogens in both captive and wild reptiles.

Surveillance and management

Wildlife Health Australia administers Australia's general wildlife health surveillance system, in partnership with government and non-government agencies. Wildlife health data is collected into a national database, the electronic Wildlife Health Information System (eWHIS). Information is reported by a variety of sources including government agencies, zoo based wildlife hospitals, sentinel veterinary clinics, universities, wildlife rehabilitators, and a range of other organisations and individuals. Targeted surveillance data is also collected by WHA. See the WHA website for more information https://wildlifehealthaustralia.com.au/Our-Work/Surveillance and https://wildlifehealthaustralia.com.au/Our-Work/Surveillance/eWHIS-Wildlife-Health-Information-System.

There are currently no targeted surveillance programs for reptile fungal diseases. There are a number of cases of reptile fungal disease reported in eWHIS. We encourage those with laboratory confirmed cases of this condition in native Australian or feral animals to submit this information to the national system for consideration for inclusion in the national database. Please contact us at admin@wildlifehealthaustralia.com.au.

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