

EXOTIC - White-nose syndrome

Fact Sheet

July 2019

Introductory statement

White-nose syndrome (WNS) is a fungal disease that has caused significant declines in insectivorous bat populations in the eastern United States and Canada. White-nose syndrome has **not** been identified in Australia. **Cavers perform a vital role in protecting Australia from WNS. It is important that cavers returning or entering Australia from overseas are aware of the risk of carrying the fungus into Australia on their clothing, footwear and caving gear and take appropriate precautions.**¹ People who come in contact with insectivorous bats in Australia should be aware of the disease and report any suspect cases.

Aetiology

The causative agent of WNS is the fungus *Pseudogymnoascus destructans* (formerly *Geomyces destructans*). The fungal genome sequence is available in Genbank (Accession KV441386).

Natural hosts

Twelve cave dwelling bat species (ten *Myotis*, one *Perimyotis* and one *Eptesicus*) have been impacted by WNS in North America with the fungus found on a further eight (four *Corynorhinus*, one *Myotis*, one *Lasionycteris*, one *Lasiurus* and one *Tadarida*), with no clinical signs^[1]. In Europe the fungus has been found on eight species of *Myotis*, and one each in the genus *Eptesicus*, *Barbastella*, *Miniopterus*, *Plecotus* and *Rhinolophus*^[2], but without the mass mortalities observed in North America^[3]. Six species of bat in China (three *Myotis*, two *Murina* and one *Rhinolophus*) have also been found infected with *P. destructans*^[4].

World distribution

To date, deaths attributed to WNS have been confirmed in bats in the USA, Canada and Czech Republic. The disease was first recognised in New York State in 2006 and since then has spread along the eastern seaboard, involving 33 states, and appearing in Washington State in March 2016, an apparent 2,000 km jump from the previous westernmost detection of *P. destructans*. The fungus, but not the disease, has also been found in Mississippi, Texas, Wyoming, North Dakota and California. The disease was first identified in Canada in March 2010 in Ontario and has since spread to seven other provinces.

¹ <http://agriculture.gov.au/pests-diseases-weeds/animal/white-nose-syndrome>

There are no confirmed reports of *P. destructans* in Europe prior to 2008. However, anecdotal reports indicate the fungus may have been present as far back as the 1970s. Current surveys have found the fungus on bats throughout Europe with no gross evidence of infection and also on bats with clinical signs of WNS. Small numbers of bats were found dead in poor condition in the Czech Republic. The deaths were confirmed histologically to be associated with WNS. However, population numbers remain stable. A survey of 367 bats in Sweden failed to find any evidence of the fungus [3, 5-9]. A survey conducted in north-eastern China in 2014 and 2015 found *P. destructans* on six species of bats, and on nine of 12 cave surfaces sampled [4].

Occurrences in Australia

There have been **no** reports of *P. destructans* in Australia. A number of bats have been submitted for exclusion testing for WNS.

From 2015 to 2017, 325 live southern (*Miniopterus orianae bassanii*) and eastern bent-winged bats (*M. orianae oceanensis*) and 30 environmental samples from South Australia and Victoria were tested by PCR for *P. destructans*. All samples were negative [10].

Epidemiology

P. destructans is psychrophilic, meaning it grows best at low ambient temperatures. *In vitro* studies have found that optimal temperatures for growth are between 12.5 and 15.8 °C, with cessation of growth above 20 °C [11]. While the fungus grows best at humidity levels above 90% it is able to survive prolonged periods of low humidity, and is capable of growth on a range of environmental substrates [12].

Transmission is by direct contact from infected bats to healthy bats and by direct contact between bats and the cave substrate. The worst affected bat species tend to cluster tightly together in large colonies, thus facilitating fungal spread [13]. Airborne transmission has not been demonstrated [14]. Humans have also been implicated in the spread of the disease.

Viable *P. destructans* spores have been found on the walls of hibernation sites in Europe, while a US study detected *P. destructans* in sediment from two mines that had been closed to bats for one to two years, indicating a potential for the organism to persist in the environment [3, 15]. Ectoparasites have been considered as possible mechanical vectors of *P. destructans* and the fungus has been found on bat mites [16].

WNS is a seasonal disease with lesions first appearing in North American bats in late September, and mortalities starting in the middle of winter in January, approximately 120 days after the bats have entered hibernation. Hibernating bats have a reduced metabolic rate and immune capability rendering them more susceptible to infection and disease. Mortality peaks in March and declines during April and May, before the disease disappears post hibernation, in June [14]. An estimated one million hibernating bats have died, with populations in some hibernacula decreasing by 90 to 100%. WNS can thus have a significant impact on bat populations.

In North American bats infection is often associated with abnormal behaviour such as increased arousal from hibernation, increased grooming behaviour and flying during the day which, in turn, has resulted in mass mortalities ^[12]. Increased arousal frequency consumes additional energy reserves, so affected bats have little or no identifiable fat stores. Wing damage results in increased evaporative water loss leading to electrolyte depletion and dehydration (Reichard and Kunz 2009; Cryan et al. 2010; Warnecke et al. 2012; Warnecke et al. 2013; Cryan et al. 2013).

The bat immune system appears to be suppressed during hibernation. However, a neutrophil response does occur after the bat's body temperature has returned to euthermic levels when it emerges from hibernation. If the bat has survived the hibernation period, this immune response can be sufficient to clear the fungus from the bat's system ^[17].

In stark contrast to the situation in North America, WNS in European bats is not associated with any increases in mortality ^[3]. The reasons for this are not entirely clear but likely related to a number of factors including species susceptibility, with infected North American bats carrying significantly higher fungal loads than their European counterparts ^[2]. European bats also tend to hibernate in small clusters, rather than in large aggregations, which could partly explain the decreased effect of the fungus on these populations ^[13].

The current hypothesis is that *P. destructans* was imported into North America from Europe, possibly on shoes or equipment used in caves, which would explain the apparent increased susceptibility of North American bats to the disease. Evidence for this theory includes the initial appearance of the disease at a single site followed by radial spread, and genetic comparisons between American and European isolates ^[18,19]. Research has also found that North American bats died whether they were experimentally infected with the North American or the European strain of *P. destructans*, strengthening the theory that *P. destructans* is an invasive species from Europe ^[20].

However, the impact that the disease will have on a given bat population is governed by a number of factors, only one of which is host resistance. These include roosting/hibernation site (WNS only affects cave dwelling bats), environmental temperature, food and water availability, and bat behaviour (Holz et al. 2016).

While the large-scale mortalities seen in North America are considered less likely to occur in Australia due to its milder climate, bat species living in southern Australia could still be affected, particularly the critically endangered southern bent-winged bat, due to the additive effect of existing threatening processes, and the fact that the entire population lives within the preferred temperature zone of *P. destructans* (Holz et al. 2016).

Clinical signs

Many, but not all, affected bats have a grossly visible white or grey fungal growth on muzzles, ears and wing membranes, which can lead to scarring and necrosis. Bats may have reduced fat stores and be clinically dehydrated. Affected wings may become thinner, discoloured, have a flaky appearance and develop erosions and ulcers. Folded surfaces of severely affected wing membranes adhere to each other, tear easily, and appear to lose tone, tensile strength and elasticity ^[21]. If bats

survive the infection they are capable of healing their wing membranes, which retain a variable amount of post-WNS scarring [22].

Diagnosis

Although the appearance of white fungal material on the muzzle, ears and wings is suggestive of WNS, it is not pathognomonic as bats in the UK displayed similar lesions that were caused by *Rhizopus* and *Paecilomyces* [23]. Histopathology is considered to be the “gold standard” to confirm a diagnosis [24]. Fungal culture may also be used to confirm the presence of *P. destructans* (See Gargas et al. 2009 [25] for full description). PCR tests have been developed to detect fungus on bat wing tissue [26-28]. *P. destructans* must be distinguished from other fungal species in the same or closely related genera, which may occur in cave environments.

Ultraviolet light (366-385 nm) has been used to screen bats for WNS but is not recommended for confirmation or exclusion of WNS. Of 80 bats that were positive histologically, 79 bats showed areas of orange yellow wing fluorescence when subjected to ultraviolet light. All 88 bats that did not fluoresce were also histologically negative for WNS [29].

Pathology

Grossly, apart from the presence of white fungal material on the face and wings, affected bats may have patches of rough skin, pinpoint white foci on the muzzle, contraction of the wing membrane and a loss of pigmentation.

Microscopically, non-pigmented, branching, septate fungal hyphae with distinctive asymmetrically curved conidia cover the epidermis leading to characteristic cup-like erosions, ulceration and wing membrane infarction with minimal evidence of inflammation [21, 24, 30]. There may be minimal inflammatory response in affected hibernating bats.

Differential diagnoses

Other superficial fungal diseases may present with similar clinical signs. In Australia, diagnoses in bats that have been investigated for WNS include overgrowth of saprophytic fungi, mite infestation, and infection with bacteria or other fungi [31-33].

Laboratory diagnostic specimens

To submit samples for exclusion testing please contact your local WHA Coordinator to discuss arrangements for processing samples.

Members of the public should not handle bats. If you find an injured or sick bat, contact a wildlife care organisation or your local veterinarian. People trained in the handling of bats should have current rabies immunity (vaccination) and always use appropriate protection when interacting with bats. Details about procedures for handling bats and national guidelines on pre-exposure prophylaxis for lyssavirus are available on the Communicable Diseases Network Australia website:

www.health.gov.au/internet/main/publishing.nsf/Content/cdnasongs.htm See Rabies and ABLV SoNG.

Wearing a filtration face mask in caves and when handling bats has also been recommended (Rose pers. comm.).

National guidelines for sample submission for WNS are available on the Wildlife Health Australia website:

www.wildlifehealthaustralia.com.au/Portals/0/Documents/ProgramProjects/National_Guidelines_for_Sample_Submission_WNS_Exclusion_Testing.pdf.

In summary: carcasses, tissues samples and/or swabs of affected skin can be collected into a sterile container for direct microscopy, molecular testing and culture ^[12, 25]. Contaminating fungi can grow opportunistically within samples very quickly, so keeping the **samples at 4 °C, prompt submission and ensuring the receiver is ready to receive the samples are essential.**

Tissues should be collected in formalin for histological evaluation. Sticky tape preparations are the least desirable samples due to artefacts and the inability to use the specimen for culture or PCR.

Photos of gross lesions should be taken prior to sampling and sent with samples. Full details of the sampling event should be recorded and submitted (see National sampling guidelines).

Treatment

There is currently no treatment for WNS. Bats may recover from the disease and clear the fungus if supported through the winter period by bringing them into captivity, raising their body temperature and providing food ^[34].

A recent study found that disease progression was unaffected by the topical application of apple cider vinegar. However, when affected little brown bats were warmed to between 18.3 °C and 23.9 °C, administered lactated Ringer's solution subcutaneously and fed mealworms, 25 out of 26 individuals recovered from the disease and were PCR negative for the fungus 70 days after being brought into captivity ^[17].

Certain volatile organic compounds, produced by bacteria, have fungistatic activity, potentially giving them a role as chemical control agents. Decanal, 2-ethyl-1-hexanol, nonanal, benzothiazole, benzaldehyde and N,N-dimethyloctamine all inhibited the growth of *P. destructans* in experimental studies ^[35].

Pseudomonas spp. and *Rhodococcus rhodochrous* have also shown an ability to inhibit growth of *P. destructans* in *in vitro* experiments ^[36, 37]. *Candida albicans* excretes trans, trans-farnesol, a bioactive sesquiterpene that has been shown to kill several fungal species. *In vitro* studies have shown that *tt*-farnesol inhibits conidial germination and hyphal growth of *P. destructans* ^[38].

Antifungal testing showed that *P. destructans* was susceptible to amphotericin B, ketoconazole, itraconazole, posaconazole and voriconazole. It was resistant to flucytosine, caspofungin, anidulafungin and micafungin and had dose dependent sensitivity to fluconazole [39].

Prevention and control

Wildlife Health Australia, in consultation with stakeholders, developed response guidelines to assist relevant agencies should the disease appear in Australia:

https://wildlifehealthaustralia.com.au/Portals/0/ResourceCentre/BatHealth/WNS_response_guidelines_1.1_Jul_2019.pdf. A range of response options are outlined in the guidelines. The preferred options will depend on the situation but may include a combination of activities to prevent further WNS transmission by humans and bats, surveillance to detect the extent of the disease, communication and education to assist with early detection and prevention of spread, and support for infected bats and bat populations.

Because of the possibility of human spread, biosecurity and decontamination would be critical in the event of an outbreak, to prevent transfer of pathogen on clothes, boots, equipment and other fomites.

The US National White-Nose Syndrome Decontamination Protocol (www.whitenosesyndrome.org/topics/decontamination) and the Canadian Wildlife Health Cooperative National White-nose Syndrome Decontamination Protocol (www.cwhc-rcsf.ca/wns_resources.php) are detailed and regularly updated.

Dedicated clothing and equipment should be used for infected caves, and ideally for all sites. Where decontamination is required, the items should first be cleaned of all mud and debris. Clothing and other suitable items should then be submersed in hot water maintained at a temperature of at least 55 °C for a minimum of 20 minutes. Equipment that cannot be immersed in water can be treated by disinfection. Disinfectants that have been shown to kill *P. destructans* include chlorine bleach, 60% ethanol, 60% isopropanol and 3% hydrogen peroxide
www.whitenosesyndrome.org/topics/decontamination.

It is important that covers returning or entering Australia from overseas be aware of the risk of carrying the fungus into Australia on their clothing, footwear and caving gear and take appropriate precautions to disinfect their equipment and clothing prior to entry into the country. People who come in contact with insectivorous bats in Australia should be aware of the disease and report any suspect cases.

Information on how to recognise and report a suspect case of WNS is available on the WHA website:

https://wildlifehealthaustralia.com.au/Portals/0/ResourceCentre/BatHealth/How_to_report_a_suspect_case_of_WNS.pdf.

Veterinarians and scientists sampling bats for WNS exclusion should use appropriate PPE and decontamination protocols. Any bat where WNS is suspected should be kept separately and isolated from all other bats and animals to reduce the risk of disease transmission.

The U.S. Fish and Wildlife Service has written “A National Plan for Assisting States, Federal Agencies, and Tribes in Managing White-Nose Syndrome in Bats”, May 2011 (www.whitenosesyndrome.org/static-page/planning-our-response).

Surveillance and management

There have been no reports of WNS in any bats from Australia's States and Territories. Suspect cases should be reported to your local WHA Coordinator (www.wildlifehealthaustralia.com.au). WHA is interested in receiving reports of any testing or field observation of the health of bat colonies from anyone working in the field with these animals. Contact us at admin@wildlifehealthaustralia.com.au.

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 no reports of WNS in Australian bats in the National Wildlife Health Surveillance Database.

Research

Current overseas research focuses on surveying caves/mines to identify new WNS-affected sites and track known sites; predicting the potential for future WNS spread; investigating biological or chemical treatment and control strategies; determining if there is resistance to WNS among bat populations; and developing a better overall understanding of the disease.

Some surveys of at-risk bat groups have been conducted, to determine if the fungus is present in Australia ^[10]. A qualitative risk assessment examining the possible introduction of WNS into Australia and its potential consequences has been undertaken ^[40].

Knowledge gaps in relation to WNS risk in Australia include fungal species present in Australian caves; temperature and humidity in Australian caves; body temperature of Australian bats during torpor/hibernation; susceptibility of Australian microbats to WNS; population and ecology of at-risk bat species; and the economic importance of insectivorous bats to agriculture in Australia.

Human health implications

No direct human health risk from WNS has been identified. There is no information indicating that people or other animals have been affected after exposure to the fungus. However, people handling bats should use safe work practices and personal protective equipment (PPE).

There is a risk of exposure through handling bats to other diseases such as Australian bat lyssavirus (ABLV).

Conclusions

White-nose syndrome (WNS) is an emerging fungal disease that has caused significant declines in insectivorous bat populations in the eastern United States and Canada. It has not been identified in Australia. A risk assessment concluded that there is a high likelihood that *P. destructans* will enter Australia in the future. Although the scale of mortalities is not expected to be as severe as in America, significant impacts may still occur, particularly for already threatened populations. The loss of bats would also have a broader impact on the ecosystem. Prevention and preparedness activities have occurred in Australia, including development of response guidelines to assist in the event of an outbreak in Australia. To assist with early detection, any suspect case of WNS in an Australian microbat should be reported.

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Wildlife Health Australia recognises the Traditional Custodians of Country throughout Australia. We respectfully acknowledge Aboriginal and Torres Strait Islander peoples' continuing connection to land, sea, wildlife and community. We pay our respects to them and their cultures, and to their Elders past and present.

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Further information on WNS is available through the Australian Government Department of Agriculture and Water Resources website (<http://agriculture.gov.au/pests-diseases-weeds/animal/white-nose-syndrome>), the USGS National Wildlife Health Center website (www.nwhc.usgs.gov/disease_information/white-nose_syndrome/) and the North American website White-noseSyndrome.org - A Coordinated Response to the Devastating Bat Disease (www.whitenosesyndrome.org).

See Holz et al. (2016) for a detailed summary of WNS as it relates to the Australian situation.

ProMed postings on WNS can be found at:

<http://www.promedmail.org/pls/otn/f?p=2400:1201:2083370501407005>.

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