Amping up Biosecurity for Herps

SIMPLE, STRICTER PRACTICES CAN HELP PROTECT REPTILES AND AMPHIBIANS FROM DISEASE

By Molly C. Bletz, Jenna Palmisano, James T. Julian, Lisa Shender and Deanna H. Olson

As amphibians and turtles top lists of species with conservation concerns, the lens of wildlife management is increasingly focused on measures to reduce threats to them at local, regional and national levels.

Reptiles and amphibians face a mosaic of risks—climate change, habitat degradation, the pet trade and overexploitation among them. Yet diseases pose some of the greatest threats to their populations. Pathogens and parasites continue to arise, jeopardizing these diverse animals, which are so central to food webs and ecosystem integrity.

Credit: Kim Titterington/Swamp Girl Adventures Reptile Rehabilitation

▼ **Jenna Palmisano wears gloves when handling pygmy rattlesnakes (*Sistrurus miliarius*) in Florida to reduce human-mediated transmission of both the snake fungal disease ophidiomycosis, caused by the skin fungus *Ophidiomyces ophidiicola*, and pentastomiasis, caused by the lung pentastome *Raillietiella orientalis*.**

In North America, biosecurity practices are being more widely adopted to reduce humans' role in transmitting amphibian disease—both between animals and between sites. We need to amp up these practices. Using higher disinfectant concentrations and longer disinfectant contact times can help combat amphibian diseases. Increasing disposable glove usage and disinfection procedures can reduce the spread of reptile diseases.

Protecting amphibians

In the 1990s, the amphibian skin disease *Bd* chytridiomycosis—caused by the aquatic fungus *Batrachochytrium dendrobatidis*—was first described. While it remains a concern for some amphibian populations, increased attention is being given to its newfound relative, *B. salamandrivorans*, or *Bsal*.

Like *Bd*, *Bsal* is thought to be native to Asia. It emerged in Europe just over a decade ago, where it continues to cause salamander die-offs. It has yet to be documented in North America, but most experts feel it is only a matter of time before *Bsal* finds a back door to our continent.

Because North America is a world salamander biodiversity hotspot, if *Bsal* reaches here, we can likely expect widespread effects (Gray et al. 2023). Lines of defense are already conceived, but additional updates are needed. To reduce human-mediated transmission of invasive disease-causing pathogens of native wildlife, many field crews engage in standard biosecurity practices that include disinfecting field gear and equipment both within a site and between sites (Gray et al. 2017; Julian et al. 2020; Olson et al. 2021).

These standard practices do not go far enough, though. Higher disinfectant concentrations are needed to prevent *Bsal* transmission (Van Rooij et al. 2017).

Bsal disinfection

Disinfectants can be a powerful tool in minimizing pathogen spread. However, they must be used at proper concentrations, applied with suitable contact times and appropriately stored so that efficacy is not reduced via biodegradation.

We echo recently revised field-disinfection procedures to emphasize higher disinfectant concentrations for *Bsal* (NE PARC 2022; Bletz et al. 2023). Virkon, bleach and ethanol can all be used. Each has advantages in particular contexts. Ethanol, for example, is useful for disinfecting delicate equipment. Virkon Aquatic is a particularly suitable disinfectant for field gear and larger equipment due to its reduced corrosiveness and toxicity. It can be prepared by adding 1.3 ounces of powder to 1 gallon of water, but care is needed, as the powder should not be inhaled. This requires working in a well-ventilated area and/or using face masks.

If store-bought liquid bleach is used to disinfect field gear, note that concentrations of the active ingredient, hypochlorite, differ in these products and can be between 5.25% and 8%.

For store-bought bleach with an 8% hypochlorite concentration, add one part bleach to four parts water (for a gallon of water, add four cups of bleach). Allow the concentration to contact the items being disinfected for five minutes, then rinse thoroughly with water. Online bleach calculators are useful tools for preparing appropriate bleach solutions. One we use is www.omnicalculator.com/chemistry/bleach-dilution.

It is inadvisable to use containers of store-bought bleach that have been opened for a month or more because hypochlorite is not stable and may not be effective. Light will also degrade bleach solutions that are stored in transparent containers. The revised bleach disinfectant concentration for *Bsal*—1.6%, or 16,000 parts per million hypochlorite—is effective for other wildlife disease pathogens, including *Bd*, ranavirus pathogens and *Ophidiomyces ophidiicola*. Follow product label instructions for use and disposal of these disinfection chemicals.

Credit: Northeast PARC

▲ **Field gear disinfection process for amphibians and reptiles: A) Secure each animal (here, a salamander is secured in a new disposable bag) and use new gloves for handling. B) Acquire equipment used for disinfection. C) Scrub debris off gear, then rinse with water prior to applying disinfectant. D) Apply disinfectant and follow product instructions for proper contact time. E) Rinse off disinfectant with clean water (From NE PARC 2022).**

Reptile worries

Concerns for North American reptile disease biosecurity are rising as reptile disease reports increase. Some of these diseases have human-mediated transmission and ties to invasive pathogens from other regions of the world. Three fungal foes warrant mention.

First, recent evidence supports the hypothesis that the snake fungal pathogen *Ophidiomyces ophidiicola* (*Oo*) has been introduced to the United States (Ladner et al. 2022). This pathogen causes

Forestall *Bsal* (*Batrachochytrium salamandrivorans*) with higher disinfectant concentrations, which also are effective against *Bd (B. dendrobatidis)*, *Ranavirus* spp., and *Ophidiomyces ophidiicola*. Prepared solutions indicate the final concentration of the active ingredient. (From NE PARC 2022; Bletz et al. 2023)

Virkon Aquatic®	Bleach*	Ethanol
1% solution	1.6% hypochlorite solution	70% solution
For personal gear and large equipment	For personal gear and large equipment	For delicate equipment

*Note that bleach can be sold at various concentrations. Be sure to check this and make the correct dilution.

the skin disease ophidiomycosis. Although our understanding of the disease is still developing, *Oo* is not ubiquitous across the continental U.S. It is known to occur in 26 states, mostly in the eastern U.S., as well as in Puerto Rico and Ontario, Canada. The disease has infected more than 49 native snake species and three nonnative species (Haynes and Allender 2021).

For more information, check out these resources:

- Partners in Amphibian and Reptile Conservation Disease Task Team: parcplace.org/species/parc-disease-task-team
- PARC resources: parcplace.org/resources/herpetofaunal-disease-resources
- North American Bsal Task Force: salmanderfungus.org

The higher disinfectant concentrations cited above are effective against *Oo* (see Haynes and Allender 2021). Hence field-gear disinfection between sampled individuals or sites merits consideration to forestall human-mediated *Oo* spread.

Second, additional pathogens causing skin lesions in snake and lizard species of North America are the fungi of the genera *Paranannizziopsis* (Lorch et al. 2023) and *Nannizziopsis* (Gentry et al. 2023). Little is known about these pathogen taxa in the wild, but there are several reports in captive lizards, tuataras and snakes. In Florida, nonnative wild chameleons have been found with *Paranannizziopsis* infections, a warning of the potential for pathogen spillover to native squamate populations (Claunch et al. 2023).

Third, a turtle shell fungus, *Endomyces testavorans*, has been described causing shell lesions in some North American aquatic turtles (Woodburn et al. 2019).

These three reptile fungal pathogens are a new frontier for both science advancement and management response for biosecurity and conservation actions. Hygiene measures such as disposable glove use and field-gear disinfection procedures are especially important for people handling reptiles across regions or between wild and captive populations.

Lungworm risks

In addition to the fungal diseases described above, lungworms present a concern for snakes. Snake pentastomiasis is the disease caused by a taxonomic group of parasitic crustaceans known as snake lung pentastomes. Although several are native to North America, the pentastome *Raillietiella orientalis* is native to Asia and is thought to have been introduced to the U.S. from the established population of Burmese pythons (*Python bivittatus*) in South Florida. At least 18 native Florida snake species across 20 Florida counties have been found infected with *R. orientalis*, with the observations of infections in snakes spreading northward (Miller et al. 2020; Palmisano et al. 2023).

Credits: A. Dane Conley/Virginia Tech; B. Jenna Palmisano/Univ. Central Florida; C. Jenna Palmisano/Univ. Central Florida; D. Terence Farrell/Stetson University

Biosecurity measures can reduce human-mediated transmission of emerging reptile diseases. A: A rainbow snake (*Farancia erytrogramma*) with lesions from the snake fungal disease ophidiomycosis, caused by *Ophidiomyces ophidiicola* shown at the arrows. B-D: Snake pentastomiasis caused by *Raillietiella orientalis*. In image B, the pentastome is circled on the road adjacent to road-killed southern black racer (*Coluber constrictor priapus*). In image C, an arrow points to the pentastome in a black racer's mouth (note that gloves are recommended). Image D shows pentastomes removed from the lungs of a scarlet king snake (*Lampropeltis elapsoides*) during a necropsy.

These pentastomes have a complex life history, cycling through invertebrate and vertebrate intermediate hosts before fully developing in their definitive snake hosts (Palmisano et al. 2022). With an abundance of potential intermediate hosts and the use of broad taxa of native U.S. snakes as definitive hosts, biosecurity concerns for the invasive pentastome are paramount in both wild and captive contexts.

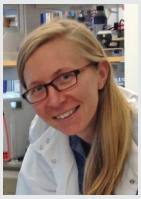
At present, *R. orientalis* infections have been found in four captive snakes, three of which died. Although it is unknown whether *R. orientalis* can infect humans, other pentastome species can. Given that potential, it is prudent to adopt the elevated biosecurity measures of glove use and gear disinfection.

Limiting our role

With a daunting potential multiple-front assault of novel disease-causing agents in North America, now is the time to amp up broader best practices for wildlife health to reduce human-mediated pathogen transmission to North American reptiles and amphibians.

Preventing pathogen spread to novel hosts is an important component of retaining North American ecosystem integrity. It's time to refresh your field biosecurity kit.

Don't be a vector. Be a wildlife protector!



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