

Salamander Chytrid Fungus (*Batrachochytrium salamandrivorans*) in the United States—Developing Research, Monitoring, and Management Strategies



Open-File Report 2015–1233

Cover. Eastern red-spotted newt, one of the species of concern should *Batrachochytrium salamandrivorans* be introduced into the United States. Photograph by Adrienne Brand, U.S. Geological Survey.

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By Evan H. Campbell Grant, Erin Muths, Rachel A. Katz, Stefano Canessa, Michael J. Adams, Jennifer R. Ballard*, Lee Berger, Cheryl J. Briggs, Jeremy Coleman, Matthew J. Gray, M. Camille Harris, Reid N. Harris, Blake Hossack, Kathryn P. Huyvaert, Jonathan E. Kolby, Karen R. Lips, Robert E. Lovich, Hamish I. McCallum, Joseph R. Mendelson III, Priya Nanjappa, Deanna H. Olson, Jenny G. Powers, Katherine L. D. Richgels, Robin E. Russell, Benedikt R. Schmidt, Annemarieke Spitzen-van der Sluijs, Mary Kay Watry, Douglas C. Woodhams, and C. LeAnn White, (*authors are listed in alphabetical order)

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Abbreviations

AFWA	Association of Fish and Wildlife Agencies
ARMI	U.S. Geological Survey, Amphibian Research and Monitoring Initiative
<i>Bsal</i>	<i>Batrachochytrium salamandrivorans</i>
<i>Bd</i>	<i>Batrachochytrium dendrobatidis</i>
DoD	U.S. Department of Defense
EPA	U.S. Environmental Protection Agency
NPS	U.S. National Park Service
NWHC	U.S. Geological Survey, National Wildlife Health Center
OIE	World Organization for Animal Health
<i>Pd</i>	<i>Pseudogymnoascus destructans</i>
U.S.	United States
U.S.C	U.S. Code
USGS	U.S. Geological Survey
USFWS	U.S. Fish and Wildlife Service
USFS	U.S. Forest Service

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Abstract

The recently (2013) identified pathogenic chytrid fungus, *Batrachochytrium salamandrivorans* (*Bsal*), poses a severe threat to the distribution and abundance of salamanders within the United States and Europe. Development of a response strategy for the potential, and likely, invasion of *Bsal* into the United States is crucial to protect global salamander biodiversity. A formal working group, led by Amphibian Research and Monitoring Initiative (ARMI) scientists from the U.S. Geological Survey (USGS) Patuxent Wildlife Research Center, Fort Collins Science Center, and Forest and Rangeland Ecosystem Science Center, was held at the USGS Powell Center for Analysis and Synthesis in Fort Collins, Colorado, United States from June 23 to June 25, 2015, to identify crucial *Bsal* research and monitoring needs that could inform conservation and management strategies for salamanders in the United States. Key findings of the workshop included the following: (1) the introduction of *Bsal* into the United States is highly probable, if not inevitable, thus requiring development of immediate short-term and long-term intervention strategies to prevent *Bsal* establishment and biodiversity decline; (2) management actions targeted towards pathogen containment may be ineffective in reducing the long-term spread of *Bsal* throughout the United States; and (3) early detection of *Bsal* through surveillance at key amphibian import locations, among high-risk wild populations, and through analysis of archived samples is necessary for developing management responses. Top research priorities during the preinvasion stage included the following: (1) deployment of qualified diagnostic methods for *Bsal* and establishment of standardized laboratory practices, (2) assessment of susceptibility for amphibian hosts

(including anurans), and (3) development and evaluation of short- and long-term pathogen intervention and management strategies. Several outcomes were achieved during the workshop, including development of an organizational structure with working groups for a *Bsal* Task Force, creation of an initial influence diagram to aid in identifying effective management actions in the face of uncertainty, and production of a list of potential management actions and key research uncertainties. Additional products under development include a *Bsal* Strategic Action plan, an emergency response plan, a monitoring and surveillance program, a standardized diagnostic approach, decision models for natural resource agencies, and a reporting database for salamander mortalities. This workshop was the first international meeting to address the threat of *Bsal* to salamander populations in the United States, with more than 30 participants from U.S. conservation and resource management agencies (U.S. Fish and Wildlife Service, U.S. Forest Service, U.S. Department of Defense, U.S. National Park Service, and Association of Fish and Wildlife Agencies) and academic research institutions in Australia, the Netherlands, Switzerland, the United Kingdom, and the United States.

Introduction

Amphibians are the most endangered group of vertebrates, with nearly a third of all species threatened with extinction globally (Stuart and others, 2004; International Union for Conservation of Nature, 2015). Although habitat loss is a major threat to species persistence, amphibians on protected lands are declining because of additional stressors, such as disease (Collins and Storer, 2003). The United States has the highest diversity of salamanders in the world (Buckley and Jetz, 2007). Of the 191 salamander species in the United States, many are endemic to only the southeastern states making the United States the global hotspot for salamander biodiversity (Wiens, 2007). Because of this, managing disease threats to U.S. salamanders is a high priority for the global conservation of salamanders.

The pathogenic chytrid fungus, *Batrachochytrium salamandrivorans* (*Bsal*), was first described in 2013 after observations of unusual mortality among wild and captive fire salamander (*Salamandra salamandra*) were reported in Europe (Spitzen-van der Sluijs and others, 2013; Martel and others, 2013). Although susceptibility to *Bsal* varies among species, the fungal pathogen is lethal to salamanders in the United States from the families Salamandridae and Plethodontidae (Martel and others, 2014). Available evidence suggests that *Bsal* is endemic to Asia, where it has been found in museum specimens more than 150 years old (Martel and others, 2014). Based on this endemicity, the discontinuity of the global incidence of *Bsal*, and the popularity of salamanders as pets, the original emergence of this pathogen in Europe is presumed to have occurred by way of the amphibian trade (Cunningham and others, 2015). More than 28 million

amphibians were imported in the United States throughout a 6-year period during the last decade (Schloegel and others, 2009; Yap and others, 2015). Thus, there is serious concern that this pathogen has already been introduced or will be introduced into the United States in the near future through the salamander trade (Cunningham and others, 2015; Yap and others, 2015). This is of particular conservation concern because the eastern United States is home to 141 species in family Plethodontidae, with 1 of 3 species tested for susceptibility in this family exhibiting mortality because of *Bsal* exposure (Martel and others, 2014). Preliminary testing also determined that *Bsal* was lethal to other genera within the family Salamandridae: the rough-skinned newt, *Taricha granulosa*; and the Eastern newt, *Notophthalmus viridescens* (Martel and others, 2014). To date, this study provides the best available data for researchers to evaluate the susceptibility of other U.S. salamander species. Given the high salamander diversity and the susceptibility of some of the few species tested, the arrival of *Bsal* to the United States could pose a considerable threat to global salamander diversity. The susceptibility of species within Salamandridae and Plethodontidae is especially concerning because many species in these families are considered relatively common. Furthermore, limited protective status of common species typically results in limited authority for management agencies to act should *Bsal* be identified or population declines observed. Evidence that North American species can become infected but survive is limited to the lesser siren, *Siren intermedia*, and Northern slimy salamander, *Plethodon glutinosus* (Martel and others, 2014), and all infected species could act as a reservoir host if the pathogen is introduced. This pathogen has the potential to have devastating consequences for U.S. salamanders, similar to extirpations observed in frog species in South America and Australia from the related fungal pathogen *Batrachochytrium dendrobatidis* (*Bd*; Lips and others, 2006; Skerratt and others, 2007).

The pace of global commerce and the accelerated emergence of new infectious diseases (that is, fungal pathogens, viruses and parasites) are increasing risk to native wildlife populations. Typically, federal and state natural resource agencies of the United States have responded to disease threats post-invasion because this is when pathogens are first detected. The identification of *Bsal* from die-offs in Europe and the documented threat this fungus poses to some U.S. salamanders provides a unique opportunity to develop control strategies for a novel infectious disease before widespread declines are observed in native populations. The U.S. Geological Survey (USGS) Amphibian Research and Monitoring Initiative (ARMI) held a workshop (23–25 June 2015) at the USGS Powell Center in Fort Collins, Colorado, USA, to initiate a proactive response (for example, identify management and research needs before invasion) for the likely *Bsal* invasion. Participants represented U.S. natural resource management agencies [U.S. Fish and Wildlife Service (FWS), U.S. Forest Service (USFS), U.S. Department of Defense (DoD), U.S. National Park Service (NPS)], and states represented by the Association of Fish and Wildlife Agencies

(AFWA), zoological parks, and academic research institutions from around the world (Australia, Netherlands, Switzerland, United States, and United Kingdom). Participants were tasked to identify research needs and potential management responses for mitigating effects of *Bsal* in the United States pre- and post-invasion. A *Bsal* invasion was framed as a crisis situation, with the United States in a precrisis stage. Precrisis and crisis stages require urgent management decisions in the face of considerable uncertainty. Participants used a decision-analytic approach (Keeney, 1996; Clemen, 1996) to explicitly articulate links between research priorities, potential management strategies, and ultimately, conservation objectives. Decision analysis also provides a template for developing an adaptive management program (Williams, Szaro, and Shapiro, 2007) in which research targets critical scientific uncertainties to improve management outcomes for *Bsal* and salamanders, as well as for the management of future novel infectious disease threats to wildlife. During the workshop, participants used several decision-analytic tools to aid in articulating the *Bsal* conservation and management problem, identifying critical scientific uncertainties and developing alternative management strategies for mitigating *Bsal* invasion and associated salamander declines in the United States.

Major Findings

Stages of *Bsal* Invasion

Pathogen invasion can be categorized into four major stages based on disease ecology and dynamics (pre-invasion, invasion front, epidemic, and establishment). These stages can be particularly useful for identifying which alternative management actions should be applied and when they can be most effective (Langwig and others, 2015). Participants agreed that these stages could be challenging to identify for *Bsal*, and many other pathogens, because information is rarely perfect. This results in uncertainty in determining pathogen presence because of imperfect detection and in defining the invasion stage because of imperfect knowledge of system dynamics. For these stages to be useful for controlling *Bsal*, uncertainty must be incorporated into the assessment management strategies. During the workshop, participants focused on developing research, monitoring, and management priorities in the context of the preinvasion stage as well as all postinvasion stages combined (invasion front, endemic, and establishment). *Bsal* has not yet been detected in the United States; therefore, many preinvasion research and management strategies focused on invasion prevention are applicable (although surveillance efforts have thus far been limited, reducing confidence that the United States is, in fact, still in the preinvasion stage). If and when *Bsal* is detected in the United States, postinvasion research and management strategies could be implemented and stages within postinvasion will be explored further.

Preinvasion

Although *Bsal* has been detected in the United Kingdom, the Netherlands, Belgium, and Germany in association with the salamander trade (Cunningham and others, 2015; Mills, 2015), *Bsal* detection has not yet been reported in the United States; thus, early detection and prevention of *Bsal* introduction into captive and wild populations are top priorities to control pathogen invasion. Because commercial trade in salamanders is considered to be the most likely pathway for entry of *Bsal* into the United States, the Lacey Act (1990; 16 U.S.C. §§ 3371–3378 and 18 U.S.C. §§ 42–43) potentially could be used to minimize the opportunities for pathogen entry by restricting salamander importation. Although the Lacey Act does not allow for listing of *Bsal* or any pathogen as an injurious species, salamander hosts of *Bsal* may qualify for listing. Listing host species may effectively reduce the number of infected animals that enter the United States while a clean-trade program that requires certification of disease-free salamander importation is being developed. Neither listing as injurious nor a clean-trade program will address potential risks from black market imports and could increase that component of risk.

Aside from the Lacey Act, there are no other regulatory tools available to address and manage wildlife diseases, which was of major concern to participants. Participants stated that preventing *Bsal* entry into the United States was crucial because there are few effective management options for *Bsal* following exposure and establishment in the wild. Efforts to restrict legal importation of salamanders from Asia and Europe through major U.S. ports could be the most effective approach to prevent *Bsal* invasion, although participants agreed that import restrictions would be unlikely to completely mitigate the risk presented by *Bsal*. Under import and trade policies, salamanders may be imported into the United States without requirement to screen animals for known diseases or to quarantine potentially sick animals. Early detection of *Bsal* in wild populations can only be accomplished through a robust, well-designed surveillance of natural populations of salamanders and newts that responds to morbidity and mortality events reported through a well-organized database and communication network. Participants predicted the introduction of *Bsal* to the United States to be certain, if not imminent, and strongly recommended exploration and development of management strategies for *Bsal* postinvasion.

Postinvasion

When *Bsal* is detected in the United States, immediate management actions to prevent the spread of the pathogen, such as restricting site-level access, decontaminating a site, and removal of amphibians from the site, could be considered. Participants, however, agreed that containment management responses would be ineffective in preventing the long-term spread of *Bsal* (that is, the invasion front may be slowed but

likely will not be halted indefinitely), especially if not acted on with urgency and decisiveness on first identification. Developing management options that aid the long-term survival of amphibian populations, despite predicted *Bsal* invasion and establishment, is of critical importance. Some pathogen and population-level management actions, such as bioaugmentation (see Harris and others, 2009), selective breeding for tolerance or resistance, and prophylactic treatments, may become available to management agencies with further research and development, but participants expressed limited confidence in their applicability and efficacy (table 1).

To assess the effectiveness of alternative management strategies, a model of the system (that is, amphibian population responses to *Bsal*) and knowledge of the ecological state of *Bsal* (that is, stage of invasion based on occurrence and prevalence of *Bsal*) are necessary. The state of the system can be identified using surveillance data collected (1) at key pet-trade import and captive salamander locations; (2) in high-risk wild populations adjacent or well-connected to potential sources of entry (see Richgels and others, unpub. data; and Yap and others, 2015); and (3) in archived swab, tissue, and extracted DNA collections. Once the state of the system is identified, management strategies at various spatial scales (that is, national, regional, and local) could be implemented to reduce disease incidence and pathogen spread into unaffected populations.

During the workshop, natural resource management agencies and researchers articulated fundamental objectives for *Bsal* and salamanders within the United States to guide the development of management strategies and research priorities. Additionally, participants explored how management responses at differing spatial scales may change depending on the state and dynamics of the system (that is, alternative hypotheses related to disease and salamander responses). Management agencies typically have multiple mission goals, including recreational, public use, economic, ecosystem services, and conservation of species, habitats, and ecosystems, which need to be jointly considered when choosing and implementing disease management strategies. By understanding the context within which natural resource managers make decisions about *Bsal*, values trade-offs and risk tolerance, and the critical uncertainties that affect research directions, the choice of management actions and conservation outcomes can be explored explicitly.

Key Uncertainties

Key uncertainties and initial research priorities were elicited from participating scientists. For each research area identified, participants estimated the cost, time, and value of reducing the uncertainty to increasing salamander persistence in response to *Bsal* invasion. Despite substantial variation among scientists in perceived importance, cost, and time required, researchers agreed that an immediate need included

the establishment of safe and effective laboratory practices for containing *Bsal*. Establishing these protocols soon would allow future research to be conducted with fewer biases and systematic errors. In this sense, research deemed prerequisite to other targeted research should receive priority. Participants with experience in disease pathology cautioned against making the assumption that this pathogen behaves similarly to other fungal pathogens. For example, in *Batrachochytrium dendrobatidis*, a related fungal pathogen causing declines in amphibians, there are multiple unique strains, and virulence varies among them (Fisher and others, 2009).

Workshop Outcomes

Organizational Structure

Despite limited time to understand the rapidly evolving state of science, policy, and legal authorities for response, several important outcomes were achieved during the workshop. The first was development of a national coordination framework using the *Bsal* Task Force. The *Bsal* Task Force contained an organizational structure and seven working groups: Surveillance and Monitoring, Database Management, Response, Decision Support, Research, Diagnostics, and Outreach/Communication (fig. 1). This organizational approach was modeled after other recent comprehensive response strategies for wildlife diseases in the United States (for example, “A National Plan for Assisting States, Federal Agencies, and Tribes in Managing White-Nose Syndrome in Bats” (Coleman and others, 2011) and “Plan for Assisting States, Federal Agencies, and Tribes in Managing Chronic Wasting Disease in Wild and Captive Cervids” (http://www.nwhc.usgs.gov/publications/fact_sheets/pdfs/cwd/cwd62602.pdf, accessed 15 July 2015)). Three important considerations follow for the organizational framework identified: (1) communication throughout the development and implementation of research and management responses is critical among and within working groups, as well as with outside entities (including the public and management agencies at all levels); (2) working groups would likely benefit by having leads from various management agencies (a lesson learned from the white-nose syndrome disease working group); and (3) representatives from diverse stakeholder agencies should make up the Executive Oversight Group. Preliminarily, membership and leadership roles in working groups were assigned (table 2), and participants were tasked with reaching out to others to broaden the scope of participation for these working groups, as well as for the membership for the Technical Advisory Committee and Executive Oversight Group.

Table 1. List of potential action categories considered for *Batrachochytrium salamandrivorans* (*Bsal*) management with their expected level of effectiveness (low, moderate, high) and level of confidence (low, moderate, high) in efficacy across participants. Expectations were elicited using the expert opinions of six groups of participants (each composed of approximately five individuals) during the workshop.

Potential action category	Expected relative effectiveness	Relative confidence in effectiveness
Containment of infected sites	Low	Low
Alter host species composition	Low	Low
Apply anti-fungal agents to salamanders	Low	High
Remove susceptible and tolerant salamanders from infected sites	Low	High
Limit site access (by humans and other vertebrates)	Low	High
Quarantine salamanders	Moderate	Low
Require health certification	Moderate	Low
Apply anti-fungal agents to habitats	Moderate	Low
Vaccinate salamanders	Moderate	Low
Apply probiotics to salamanders	Moderate	Low
Physical modification of habitat	Moderate	Moderate
Enforce fieldwork biosecurity	Moderate	High
Create assurance colonies	Moderate	High
Breed salamanders for resistance and/or tolerance	High	Low
Deploy <i>Bsal</i> zoospore removal methods	High	Low
Enact legislation that authorizes actions on wildlife pathogens	High	Low
Ban all importation of salamanders	High	Moderate
Restrict salamander trade	High	Moderate
Destroy habitats of infected sites	High	Moderate

Influence Diagram

Choosing optimal management action(s) for *Bsal* will undoubtedly require making decisions in the face of considerable uncertainty. Using tools from decision analysis, managers and researchers can assess how to best manage *Bsal* given a range of uncertainties and associated risk tolerances. Using the four stages of invasion for disease management framework outlined in Langwig and others (2015), participants created a simplified influence diagram that captured biotic and abiotic factors potentially affecting the invasion, spread, and establishment of *Bsal* in North American salamanders (fig 2). Possible management options (Appendix 1) and application strategies were then elicited using the broad range of participant expertise and experiences working with other infectious wildlife diseases, including Tasmanian devil facial tumor disease (an infectious cancer where the tumor cells themselves are the infectious agent), white-nose syndrome in bats caused by a fungal pathogen, *Pseudogymnoascus destructans* (*Pd*), and chytridiomycosis in frogs caused by a fungal pathogen closely related *Bsal*, *Batrachochytrium dendrobatidis* (*Bd*). Developing a framework for linking actions, consequences,

and objectives in a workshop setting with researchers and management agencies improved coordination among stakeholders and increased awareness of the importance in identifying the management context for different decision makers and stakeholders. Once the influence diagram is refined and updated with conditional probabilities and outcomes in a decision model (such as a Bayesian belief network; fig. 3; Marcot and others, 2006), it will assist in identifying the expected effectiveness of diverse actions and the most influential sources of uncertainty. Participants contributed to the development of a simplified example of such a network, summarizing the stochastic events that determine the arrival, spread, and establishment of the pathogen; their relations with salamander population dynamics, which of those processes would be affected by specific management actions; and the final outcomes in terms of the management objectives (figs. 2 and 3). A critical and extremely useful characteristic of the influence diagram and Bayesian belief network is that it can be altered to accommodate site or region-specific environmental and decision contexts and can be updated as additional information becomes available through research and monitoring.

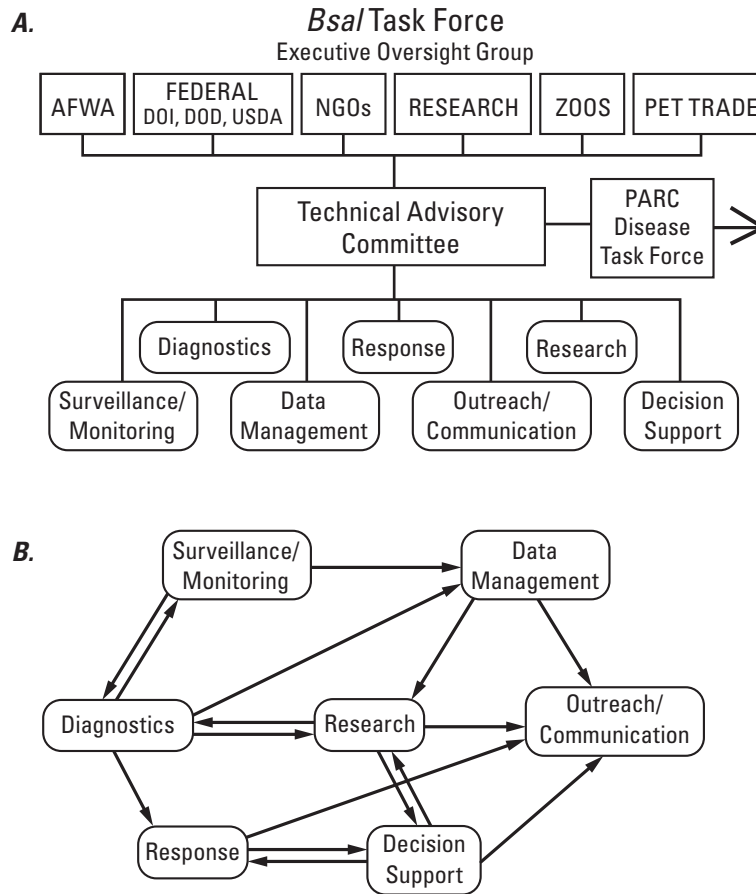


Figure 1. (A) Organizational structure for the U.S. *Bsal* Task Force with seven working groups advised by a technical advisory committee and executive oversight group composed of representatives from governmental, academic, and nonprofit organizations and (B) the flow of essential information between specific working groups (with communication occurring among all groups).

Table 2. Initial leads for each working group within the U.S. *Bsal* Task Force organizational structure. Other members will be contacted and added to each working group following the workshop.

Working group	Initial lead
Surveillance and Monitoring	Mike Adams
Diagnostics	Jenn Ballard
Research	Reid Harris
Data Management	Dede Olson
Response	Priya Nanjappa
Decision Support	Evan Grant
Outreach/Communication	Joe Mendelson

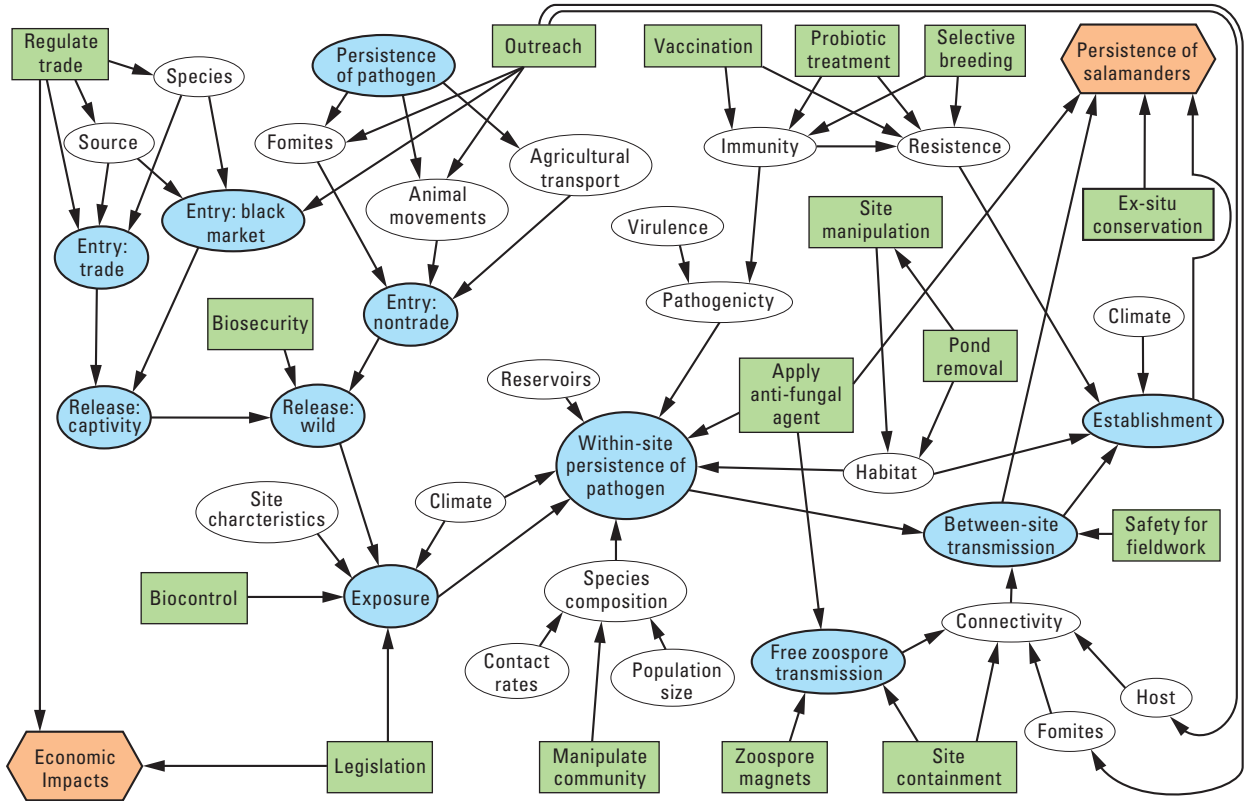


Figure 2. Simplified prototype influence diagram that links potential management actions (boxes) and abiotic and biotic factors affecting disease and amphibian processes (circles) to ultimate fundamental management objectives (hexagons; persistence of salamanders and economic impacts of management actions).

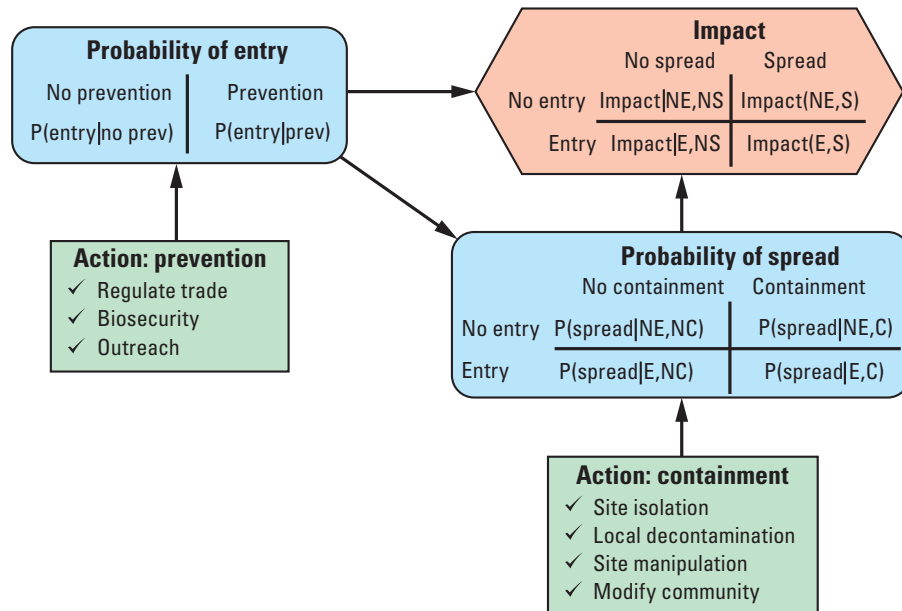


Figure 3. Example of simplified Bayesian belief network (conditional probabilities) that could be used to guide the choice of *Bsal* management actions. The outcome (impact) is conditional on stochastic events such as the entry and spread of the pathogen. Both are represented as conditional probability tables, where probabilities may or may not be affected by management actions.

Identified Urgent Needs

Response Plan for Detection

Participants urged the development of an emergency response plan to guide managers in taking action at the first detection of *Bsal* in captive or wild populations. This response plan is a component of a larger *Bsal* Strategic Action Plan (see *Bsal* Strategic Action Plan section below) but received high priority during the workshop because of the collective experience of the group with disease epidemics. Participants developed a set of flow charts that included notification and reporting recommendations, potential management actions for consideration, and additional surveillance and monitoring needs that are conditional on the circumstances and characteristics of the first, or first set, of detections. The Response Working Group (see fig. 1) will finalize this emergency response plan, which will be revised as more *Bsal* information becomes available and will be available through AFWA and the *Bsal* Task Force.

Surveillance and Monitoring Program

Pathogen and disease surveillance is essential in initiating a timely response to the likely *Bsal* pathogen invasion. Even though the detection of *Bsal* could occur from opportunistic reporting, such as submission of morbid or dead pets or encountered animals from the wild or biological supply facilities, or by sampling of archived specimens, such as those collected for *Bd*, a robust surveillance design provides the best chance for early detection and increases the options for limiting pathogen spread and its effects on host species in the United States. The Surveillance and Monitoring Working Group will design a program tailored to and based on the best available knowledge of *Bsal*. This working group will use information from two independent risk models for pathogen introduction and establishment: one model developed by Richgels and other USGS participants in the workshop (unpub. data), and another developed independently by Yap and others (2015). Although these analyses used different methods, both models identified overlapping locations of high risk for *Bsal* invasion and impact on native species. Although both models have high uncertainty, they use the best available data. An important advantage to both of the models is that new data on pathogens, host ecology, or environment can be incorporated so that surveillance strategies can be modified to take advantage of newly available information.

Participants acknowledged the importance of monitoring salamander populations in conjunction with the surveillance for *Bsal* and other important amphibian diseases (for example, *Bd* and ranavirus). Monitoring populations for pathogen occurrence and trends in population demographics can help improve risk models and prioritize where local resource managers may

want to manage salamander populations and environments before the arrival of *Bsal*. As our understanding of high-risk locations is updated, management agencies can target public outreach and engagement with stakeholders (that is, organizations and industries involved with the movement of captive and wild salamanders), as well as state agencies, Congress, and North American collaborators from Mexico and Canada.

Importantly, a surveillance and monitoring program in the absence of a management plan has limited use for controlling impacts of *Bsal* on salamander populations. Participants suggested that design of a surveillance and monitoring program would include consideration of how information gathered will be used to inform management responses. Sampling designs, therefore, may need to be developed within the decision context represented in the influence diagram (fig. 2). For example, actions for site containment (fig. 2) may require information about the following: (1) the dynamics of infection at the contained site (that is, mortality of infected individuals that could indicate a fade-out or survival of individuals that could indicate that individuals are serving as vectors), (2) the sampling protocol used at nearby sites that maximizes the probability of detecting pathogen arrival, and (3) surveys to assess animal movements from the contained site (to evaluate the effectiveness of containment). Resource constraints may limit the capacity of managers to collect all the desired data, forcing them to prioritize monitoring or to modify what circumstance will trigger an action. For example, actions that seek to modify between-site transmission could be triggered by determining pathogen presence at new sites through occupancy surveys, instead of more expensive tracking of individual salamanders, even if the former provides less information on the direction and magnitude of the transmission pathway.

Reliable Diagnostic Protocols and Database Management

Participants discussed the possibility of establishing a central testing laboratory to reduce diagnostic variability, to allow for more rapid assessment of salamander mortalities, and to facilitate evaluation of diagnostic and reporting errors. Should a central testing laboratory not be possible, participants agreed that the Diagnostic Working Group could help independent laboratories collaborate to produce reliable and standardized diagnostic protocols and practices that would be shared among participating labs. The USGS National Wildlife Health Center (NWHC) is a World Organization for Animal Health (OIE) collaborating center for research, diagnosis, and surveillance of wildlife pathogens. The NWHC has applied to be a reference laboratory for the related *Bd* pathogen and has expressed a desire to work with the Diagnostic Working Group to develop standards and serve as a primary laboratory for *Bsal* surveillance. The Diagnostic Working Group is beginning to develop standardized protocols that could be shared broadly across independent laboratories. This effort will address

concerns about the equivalency of results from multiple laboratories. Data management and accessibility are also important parts of a coordinated diagnostic effort. Participants debated several options to address this need, including adapting existing disease databases, developing a new *Bsal* database, or using the USGS All Taxa Database, and agreed to create an immediately accessible interim database while a long-term reliable data management plan is being developed.

***Bsal* Strategic Action Plan**

Participants identified the need for an overarching *Bsal* Strategic Action Plan to aid amphibian conservation given the severe threat presented by *Bsal*. The umbrella document would include a vision statement, mission, objectives, and strategies for managing *Bsal* and anticipated population declines associated with the pathogen. Specific components of the *Bsal* Strategic Action Plan were not discussed in detail during the workshop but are being developed with input from the Executive Oversight Group, Technical Advisory Committee, and each working group.

Research Priorities

Participants, using the collective experience of years of research and insights from studying wildlife disease (including other pathogenic fungi), discussed the best use of limited resources for researching and managing the likely *Bsal* invasion. The group recognized that management strategies and critical areas for research considered priority now (during the preinvasion stage of *Bsal*) might change as more information becomes available and uncertainties are reduced. Priorities discussed include the following:

1. Deployment of qualified diagnostic and detection methods for *Bsal* from swabs, tissue, preserved specimens, and environmental samples—particularly important as this information is a prerequisite to other research directions.
2. Assessment of the susceptibility in range of potential hosts, including animals other than salamanders.
3. Identification of transmission pathways to inform management control efforts, such as decontamination, quarantine, or culling.
4. Development and evaluation of the effectiveness of short-term containment measures, such as site isolation, local treatment, and methods to decontaminate sampling gear.
5. Development of long-term actions for coexistence with the pathogen, such as breeding for resistance or tolerance or both and environmental management.

Products Under Development

Participants emphasized the need for effective engagement and communication from all working groups and participating institutions as part of an effective *Bsal* Strategic Action Plan. As a result, several groups are working on standard operating procedures and information sheets (for example, the Partners in Amphibian and Reptile Conservation Disease Task Team: <http://www.parcplace.org/parcplace/images/stories/pdf/Bsal/Brief.pdf>). Additionally, participants identified key stakeholder audiences outside the *Bsal* Task Team, including state and Federal agency biologists, pet owners, zoos, and members of U.S. and State Congressional offices who will need to be kept informed as information is gathered and strategies are designed and implemented. Continual engagement and effective communication with all stakeholder groups will be essential for protecting North America's salamanders from the serious risk posed by this invasive pathogen. Other informational products are under development and include the following:

1. Article on workshop outcomes and challenges (Grant and others, unpub. data).
2. Finalized Response Plan and overarching response coordination framework and strategy.
3. Initial development of *Bsal* Strategic Action Plan.
4. White paper on research needs after the influence diagram is revised using expert elicitation techniques.
5. Decision model prototypes for specific decision makers (for example, DoD, NPS, FWS).
6. Design a surveillance and monitoring protocol program.
7. Create citizen science project on iNaturalist.org to help detect salamander mortality events (“Saving Salamanders with Citizen Science”).

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Appendix 1

Appendix 1. Initial list of potential management actions for *Batrachochytrium salamandrivorans*.

Table 1–1. Initial list of potential management actions for *Batrachochytrium salamandrivorans* (*Bsal*) generated during a brainstorming exercise and arranged into 11 broad categories: (1) environmental and habitat actions, (2) actions on animals, (3) U.S. import regulation and policies, (4) captive breeding, (5) commercial pet trade regulation and policies, (6) biological supply industry, (7) zoo industry, (8) authority and jurisdiction to act, (9) human activities, (10) monitoring and research, and (11) public education and outreach. Efficacy of each action (or combination of actions) will vary with the stage of invasion (preinvasion, invasion front, epidemic, and establishment).

(1) Environmental and habitat actions
destroy affected habitat
filter pond water
remove pond water
regulate water extraction
regulate water use
fungicide treatment to aquatic habitat
fungicide treatment to terrestrial habitat
salt addition
copper addition
heat treatment
probiotic treatment
UV treatment
zooplankton treatment
manipulate aquatic assemblage
increase habitat fragmentation
forest thinning
modify substrates
aerial application of antimycotic
zoospore magnet
abiotic-binder addition
regulate and treat wastewater
containment of site to restrict animal or pathogen dispersal
(2) Actions on animals
cull all animals
cull subset of animals
apply heat treatment
apply vaccination (topical)
apply fungicide patch
apply probiotic
apply prebiotic
expose animals to another less virulent <i>Bsal</i> strain (competition)
inject immune stimulation/booster
inject with co-infection
translocate before treatment
translocate after treatment
alter population densities

Table 1–1. Initial list of potential management actions for *Batrachochytrium salamandrivorans* (*Bsal*) generated during a brainstorming exercise and arranged into 11 broad categories: (1) environmental and habitat actions, (2) actions on animals, (3) U.S. import regulation and policies, (4) captive breeding, (5) commercial pet trade regulation and policies, (6) biological supply industry, (7) zoo industry, (8) authority and jurisdiction to act, (9) human activities, (10) monitoring and research, and (11) public education and outreach. Efficacy of each action (or combination of actions) will vary with the stage of invasion (preinvasion, invasion front, epidemic, and establishment).—Continued

(3) U.S. Import regulations and policy
ban salamander imports and enforce national import restrictions, laws, and regulations
regulate salamander imports and enforce national import restrictions, laws, and regulations
regulate and enforce state salamander import restrictions
require health certificate
quarantine imported animals
decontamination of import animals
require health certificate
require surveillance for pathogen on all traded animals
require surveillance for pathogen in water
(4) Captive breeding
create assurance colonies
selective breeding (resistance, tolerance)
population augmentation
hybridize species for low <i>Bsal</i> virulence
captive breeding with reintroduction (increase herd immunity)
genetic modification of host
genetic modification of pathogen
breed nonreproductive, highly susceptible animals to serve as fungal ‘sink’
(5) Commercial pet trade regulation and policy
require and enforce biosecurity protocols
require pre-trade health certificate
treat or decontaminate animals prior to trade; require chain of custody documentation
quarantine and test animals
voluntary pet marking
surveillance of pathogen
enforce existing salamander trade and health laws
(6) Biological supply industry
require and enforce biosecurity protocols
ban Caudata in biological supply industry
require health certificate
regulate biological supply industry
(7) Zoo industry
require and enforce biosecurity protocols
require health certificate
treat or decontaminate animals
quarantine and test animals
voluntary marking
surveillance for pathogen

Table 1–1. Initial list of potential management actions for *Batrachochytrium salamandrivorans* (*Bsal*) generated during a brainstorming exercise and arranged into 11 broad categories: (1) environmental and habitat actions, (2) actions on animals, (3) U.S. import regulation and policies, (4) captive breeding, (5) commercial pet trade regulation and policies, (6) biological supply industry, (7) zoo industry, (8) authority and jurisdiction to act, (9) human activities, (10) monitoring and research, and (11) public education and outreach. Efficacy of each action (or combination of actions) will vary with the stage of invasion (preinvasion, invasion front, epidemic, and establishment).—Continued

(8) Authority and jurisdiction to act
lobby for research and surveillance funding
improve access to actions
lobby for Federal authority to act on wildlife diseases including <i>Bsal</i>
advocate for injurious species listing
coordinate North American (Mexico and Canada) and international efforts
develop formal organizational structure, leadership, and <i>Bsal</i> coordination
engage champions (congressional, state/Federal, agency leadership)
establish political allies
maintain a cohesive <i>Bsal</i> response (staff)
engage decision makers and stakeholders from the beginning
increase public awareness (private land owners)
assess regulations for potential treatment options
(9) Human activities
reduce public access
regulate and enforce bait regulations
limit spread of other vectors (frogs)
create and enforce disinfection stations: decontamination protocols for all user groups (researchers, public, managers)
regulate and enforce collection and movement of animals (local, region, state)
(10) Monitoring and research
create and implement surveillance program
employ <i>Bsal</i> -detecting working dogs
develop rapid testing field protocols (ELISA, e-dna)
reduce diagnostic errors (centralize diagnostics, refine methodologies)
identify origin and spread of <i>Bsal</i>
update risk assessment models
develop probiotic treatments
develop vaccinations
assess susceptibility using mucosome assays
monitor effects on populations (population demography, nonlethal effects, survival)
sequence <i>Bsal</i> genome, genotypes, and strains
maintain database for information sharing
(11) Public education and outreach
outreach and communication
hand out kits for detection and eradication to landowners/agencies
increase compliance (pet trade actions)
increase compliance (local site-visit decontamination actions)
increase compliance (bait and movement of animals)
coloring books and student outreach
create a super hero (public figure/image—puppet, a voice) for salamander/ <i>Bsal</i> problem

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