

North American Fungi



Volume 12, Number 1, Pages 1-20
Published February 27, 2017

Distribution of *Heterobasidion irregulare* in Missouri pine-oak forests

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Lee, C. A., J. Bruhn, and R.-M. Muzika. 2017. Distribution of *Heterobasidion irregulare* in Missouri pine-oak forests. *North American Fungi* 12(1): 1-20. <http://dx.doi.org/10.2509/naf2017.012.001>

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Accepted for publication February 2, 2017.

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Abstract: We surveyed the Missouri Ozarks region for the pathogenic fungus *Heterobasidion irregulare*, a parasite that has gone largely disregarded within the region since the 1960s despite its potential to cause major problems for the restoration and management of shortleaf pine (*Pinus echinata*). Using spring surveys, we recovered the pathogen (as both spores and basidiomata) at 21 of 31 surveyed sites throughout the extent of the Ozarks region. Using a more intensive spore-trapping procedure, we also surveyed pure hardwood stands within the oak-pine matrix and pure hardwood forests outside the distribution of shortleaf pine. We detected some spores from pure hardwood stands that were relatively near overstory pines, but we detected no spores from pure hardwood forests at long distances (>100 m) from pines. We conducted a GIS-based analysis of pathogen presence and relative spore abundance relative to pine patch distance, pine patch size, and pine patch edge length within the surrounding

landscape and determined that distance to the nearest overstory pine as well as the number of pine patches within 500 m were important predictive variables. These data indicate that *Heterobasidion irregulare* inoculum is likely to be detected within 100 m of overstory pines in the Missouri Ozarks region and demonstrate the ability of this pathogen to persist and spread inoculum even in a landscape with significantly reduced host plant density.

Key words: shortleaf pine, pathogen, restoration, inoculum, dispersal, GIS, landscape pathology

Introduction: Forest managers recognize the basidiomycete fungal genus *Heterobasidion* as the most damaging root disease of conifers in the northern hemisphere (Hodges 1969; Woodward et al. 1998). In North America two species, *H. irregulare* Garbel. & Otrosina and *H. occidentale* Otrosina & Garbel., cause significant economic losses to managed forests. These two species also greatly affect forest stand structure, succession, nutrient cycling, and wildlife habitat in forests of all ages by creating canopy gaps, returning nutrients sequestered in woody material to the soil, and contributing downed woody material and tree cavities for animal protection and nesting (Goheen and Otrosina 1998; Garbelotto 2004).

In the lower Midwest and southeastern United States, several pine species as well as eastern redcedar (*Juniperus virginiana* L.) are susceptible to *H. irregulare* infection. Young, thinned plantation pine stands in particular suffer from large-scale mortality in this region (Alexander 1989; Filip and Morrison 1998). Despite this reputation as an economic pest, the pathogen has not been well studied in the northern part of the shortleaf pine (*Pinus echinata* Mill.) distribution in the Ozark Highlands of Missouri. The absence of research may relate to the extensive early removal of old-growth pine in the Ozarks, where loggers harvested nearly all the virgin pine resource, and where subsequent land uses encouraged colonization of the landscape by oak species (Cunningham 2007). Because of this, shortleaf pine abundance has been estimated to be somewhere between 6 and 50% of what it was at the turn of the twentieth century (Liming 1946;

Guyette et al. 2007; Scroggins et al. 2013). The USDA Forest Service produced a limited number of shortleaf pine plantations in this fragmented landscape and subsequently observed, ca. 1950s-1960s, *Heterobasidion*-caused mortality of young trees that reached up to 75% in at least one plantation (Berry and Dooling 1962). However, this wave of mortality subsided within a decade (Eng 1965; Lautz 1967), and little further attention was given to the pathogen in Missouri.

In 2013 and 2014 we conducted a non-systematic survey for *H. irregulare* in the Missouri Ozarks region, motivated by several considerations. First, the pathogen has recently increased in prominence in eastern white pine and red pine forests in Wisconsin (Stanosz et al. 1995; Erbilgin and Raffa 2002), drawing attention to gaps in our knowledge of the pathogen's distribution and ecology in mid-North America. Second, recent study of *H. irregulare* in Italy, where it is non-native and highly invasive, suggested that it unexpectedly resides in pure hardwood forests there (presumably by infecting hardwood tree species, although the host(s) have not been determined) rather than being restricted to forests containing pine; this raises the question of whether this behavior is novel in invaded habitats or is also exhibited in the pathogen's native range (Gonthier et al. 2007; Gonthier et al. 2012). Finally, a resurgence of interest in restoring shortleaf pine across native landscapes in the lower Midwest and upper South (e.g., Loewenstein and Davidson 2002; Guldin 2006; Kabrick et al. 2007; Tuttle and Houf 2007; Scroggins et al. 2013) has drawn attention to the importance of understanding the distribution of this important pathogen and its potential for

hampering such restoration efforts. These factors, plus a general lack of knowledge about the pathogen's distribution and ecology in Missouri, led to this initial effort. To accompany our general attempts to find the pathogen, we formulated the following research questions:

- How widely does *H. irregulare* occur in MO shortleaf pine forests?
- Is presence of *H. irregulare* related to nearby (within 500 m) pine patch density?
- Is presence of *H. irregulare* related to nearby (within 500 m) pine patch size?
- Is crude abundance (i.e., estimated point sporeload) of *H. irregulare* related to nearby pine patch density?
- Is crude abundance of *H. irregulare* related to nearby pine patch size?
- Is current presence of *H. irregulare* related to historic pine presence?

Materials and Methods

Study Sites. We surveyed 31 sites throughout the Missouri Ozarks and surrounding region to assess *H. irregulare* distribution, spanning several ecological subsections delineated by Nigh and Schroeder (2002) (Fig.1). To address the relationship of *H. irregulare* to hardwood and historic pine habitat in Missouri, we sampled selected pure-hardwood stands within the historic (early 19th c.) native distribution of shortleaf pine as well as selected hardwood stands 36 and 128 km outside this distribution as delineated in Harlan (2008) (Fig. 2). The sites outside of the historic distribution of pine helped us to ascertain whether *H. irregulare* commonly infects eastern redcedar in areas outside the range of pine; such infection was recently reported (S. Wright, Missouri Department of Conservation, personal communication).

These sites vary in mean temperature and precipitation throughout the year but generally exhibit a continental climate, with a mean January low temperature of -4°C and a mean July

high temperature of 32°C (Decker 2015). Precipitation averages ~1300 mm per year in the Ozarks region (Decker 2015). Topography varies greatly, from hardwood-dominated flats near the Missouri River at the northern edge of the Ozarks in Boone County to extremely dissected landscapes associated with major Ozark drainages such as the Current, Black, Gasconade, Meramec, and White Rivers. Shortleaf pine stands generally reach their greatest density and productivity on substrates weathered from the Roubidoux formation, which provides sandstone-derived acidity to the soils; areas underlain by limestone and dolomite historically have featured more mixed hardwood-pine forests (Kabrick et al. 2004; Guyette et al. 2007).

Survey Protocols. To find *H. irregulare* and estimate its relative abundance, we employed three kinds of surveying procedures of varying complexity across this landscape. Each of the first two survey procedures was centered on an approximately 60 m X 60 m area, but the second also effectively reached beyond this area through sampling of the air spora. First, we explored suspect sites for *H. irregulare* basidiomata, focusing our search on the most likely places for basidiome production: standing dead trees, windthrown trees, stumps, and logs. Second, we adapted the procedure of Gonthier et al. (2007) to detect airborne basidiospores of *H. irregulare*. For this procedure, we harvested small shortleaf pine trees from non-native habitat (Baskett Research and Education Area, Ashland, Missouri), removed the bark, and cut them into 1-cm-thick disks. Within 24 hours of cutting disks, we placed them at each survey site, with one disk in the center surrounded by four disks 10 m (at our initial sites) or 30 m (at most sites) distant in the four cardinal directions. We sprayed each disk with ethanol and placed it on the forest floor in an open petri dish upon filter paper moistened with 3 mL distilled water. We also placed a control disk at the center of each plot; this disk was identical to the others except that the lid of the petri dish was closed. This enabled us to

ascertain whether bait stock was infected with *H. irregulare* prior to the surveys. All surveys took place in March–April 2013 and/or March–April 2014, following Drummond and Bretz (1967), who determined this to be the time of maximum airborne inoculum production for *H. irregulare* in Missouri.

We exposed each disk for approximately 24 hours (range: 17.5–24 hr), retrieved it, remoistened the filter paper, and incubated it at room temperature for 7–14 days. Beginning at day 7, we searched the entire surface of each disk daily with a dissecting microscope to mark the emergence and growth of the distinctive conidiophores of *Spiniger meineckellus*, the anamorph of *Heterobasidion* spp (Stalpers 1974). We marked the perimeter of each colony with a marker or pencil to track our observations and continued the observations until day 14, at which point we counted the number of colonies on each disk, measured the diameter of the disk in two orthogonal directions to obtain an average diameter, and calculated disk area and colony-forming unit (cfu) deposition rate (cfu \cdot cm² \cdot hr). We expressed relative abundance using these rates to avoid the assumption that each observed cfu indicated one basidiospore, as some fraction of basidiospores could be expected not to germinate because of unfavorable ambient conditions, competition from other microorganisms, or other reasons.

Third, we conducted a more detailed survey at six sites (Clearwater 1, Clifty Creek CA, Compton Hollow CA, Hinkson, Fortune Hollow 2 and 3) to provide more surface area for spore capture in areas, such as pure hardwood forests, where *H. irregulare* might be expected to reside at low densities, and also to determine whether we could associate patterns of spore deposition at the plot level in these non-pine sites with the distribution of stumps and redcedar saplings or trees (the two most likely local pathogen refugia). For this survey, we used 30m X 30m plots, deploying 30 disks in each plot by dividing the

plot into a 1m X 1m grid and generating a random distribution of 30 points across this grid using the *genRandomClust* function within package ‘clusterGeneration’ of R (Qiu and Joe, 2015). We then recorded the coordinates of each stump or redcedar sapling within the plot and exposed, collected, incubated, and observed the disks as described previously.

Data Analysis: Pathogen presence. We used GIS to derive a set of predictor variables describing conifer patch characteristics in the landscape surrounding each survey center point. To derive these variables, we used a set of georeferenced aerial photographs taken at 0.61 m (2 ft) resolution during leaf-off (winter) and archived at the Missouri Spatial Data Information System at the University of Missouri. Since these photographs were taken during leaf-off, and since only two conifer species were present on the landscape, we could clearly delineate conifer patches on the imagery. Upon each photo we generated a supervised classification in ArcGIS 10.2.2, using 3–7 training classification polygons depending on the complexity of the landscape, and then from these classifications extracted circular areas with 200 and 500 m radii extending from each plot center (Fig. 3). We then used the Patch Analyst extension (Rempel et al. 2012) in ArcGIS to calculate pine cover (m²), mean patch size (ha), total edge of pine patches (m), and number of pine patches within the circular areas. Following Turner et al. (2001), we define a “patch” as an area on the landscape that differs from its surroundings in appearance; in this specific context, each patch is an area (i.e., a polygon) within the aerial photograph of similar pixel values, completely surrounded by an area of unlike values. Additionally, in the field we measured distances (m) to nearest overstory pines and then confirmed or adjusted them using the aerial photography. We used these pine patch and distance measures as independent variables to model pathogen presence or absence on the landscape, with our observations as a response variable distributed in a binomial manner

(presence=1, absence=0). We constructed logistic regression models using the *glmer* function in the *nlme* package in R (Pinheiro et al. 2014; R Development Core Team 2008) so that we could specify plot as a random effect and errors to follow a Bernoulli distribution (Pacala et al. 1993). Preliminary analysis showed pine cover and total edge to be highly correlated with the other patch measurements, so we discarded those variables for further analysis. We used a comparison of corrected Akaike Information Criterion (AICc) weights to select the most plausible model (Burnham and Anderson 2002). Additionally, we computed an R^2 for the best model using the *arm* package in R (Gelman and Su 2015) as recommended by Nakagawa and Schielzeth (2012).

Pathogen abundance. We related crude pathogen abundance, estimated as cfu deposition rate at the traps averaged at the plot level, to the same suite of predictor variables. For this analysis we used generalized linear mixed-effects models because the count response variable (cfu deposition rate) was distributed non-normally; we compared model fits under both Poisson and negative binomial distributions (using the *glmer* function for the former and the *glmmadmb* function in the R package of the same name for negative binomial mixed modeling). We used a similar model selection procedure involving AICc comparison as with the former analysis.

Spore deposition and pine stump locations. Of the six sites at which we placed 30 randomly distributed disks, we detected spores at only one (Clearwater 2). At this site spores were deposited on 9/30 disks. No redcedar existed on this plot. We mapped the stumps and calculated the distance between each disk and each stump using the *spatstat* package in R (Baddeley and Turner 2005), then summed the distance from each disk to all stumps. This was to capture the possible effect of any surviving inoculum, which would likely be located only in few, and probably randomly distributed, stumps. We then plotted

spore deposition rates against the summed distances to examine the data for patterns and used *spatstat* to produce spatial distance maps that showed graphically the distances between stumps and positive disks.

Results

Pathogen presence. We detected spores of *H. irregulare*/*S. meineckellus* or found *H. irregulare* basidiomata at 21 of 31 surveyed sites (Table 1). We recovered spores at 15, basidiomata at 11, and both at 5 of these sites. The sites varied in forest structure and characteristics, especially in the relative proportions of pine cover in the surrounding landscape. The ratio of conifer cover to hardwood cover within a 500-m-radius circle surrounding each sampling point ranged from 0.000705 (at Fortune Hollow 3) to 0.681016 (at Big Piney B). We only found one basidiome on one standing tree, attached to the roots of a shaded-out sapling at the Clearwater 1 site. All other basidiomata had either grown from stumps or on the undersides of windthrown tree trunks (Fig. 4). Windthrow damage at most sites was minor and mostly limited to isolated group windthrows of 4-5 pines, with the exception of the Sinkin 1 site, where a 2009 derecho that extended across most of the width of the state uprooted many trees of all species.

Because we detected no *H. irregulare* at the Hinkson, Clifty Creek, and Compton Hollow sites (based on surveying with 30 disks per site) and these sites lie outside the historical range of shortleaf pine, we omitted them from further analyses. No statistical differences between pine patch characteristics within 200- or 500-m-radius circles around the plot were detected (i.e., these characteristics remained proportional from smaller to larger circular areas), so we used the 500-m-radius landscape classifications for further analysis. To predict the probability of detecting the pathogen at a given site, AICc score comparison identified one best variable: distance from the nearest overstory pine (Table 2), which

is supported by the failure to detect spores on any baits at distances larger than 91 m from the nearest such pine. This model had a marginal (i.e., fixed-effects) R^2 of 0.64. Estimated random effects were so small (4.96×10^{-15}) in comparison to residual variance as to be negligible.

Pathogen abundance. We detected spores at 15 of the 31 visited sites. Models resulting from assuming a Poisson distribution for pathogen abundance count data were overdispersed, suggesting that negative binomial regression was more appropriate. Modeling of the count data using the negative binomial distribution indicated near-equal support for three possible models, all containing terms for both distance from the nearest overstory pine and the number of pine patches within 500 m from plot center (Table 3). The presence of the interaction term indicates a slight synergistic effect between the two variables: the slope of the regression increases with a combination of decreased distance from pines and increased numbers of pine patches within a 500 m radius.

Relationship of spore deposition to stump presence. We detected no spores at locations where we deployed traps (30 at each site) with redcedar present but outside of the range of shortleaf pine (Hinkson, Clifty Creek CA, and Compton Hollow CA) or at sites with only hardwood stumps onsite and shortleaf pine 100-200 m distant (Fortune Hollow 2 and Fortune Hollow 3). We detected spores on 9 of 30 disks at Clearwater 2, a site under a pure hardwood canopy but containing numerous very old pine stumps and with mature pine trees 77 m distant. Estimates of deposition rates on these 9 traps ranged from 10-74 cfu m^{-2} -hr, but these rates were not normally distributed across the positive traps. Examination of distance maps of stump concentration and positive traps did not indicate an obvious relationship between spore load and the spatial array of stumps; in fact, positive traps appeared to be clustered closest to the area of the plot with the lowest density of stumps. Plotting

the distribution of spore detections relative to the summed distances from each trap to all plot stumps confirmed this (Fig. 5). Therefore, we did not attempt statistical hypothesis testing to address the question of the relationship between cfu deposition and stump distribution in this plot.

Discussion

Heterobasidion distribution in Missouri. We used this survey to examine this important pathogen's presence in Missouri shortleaf pine forests for the first time since the 1960s and found it to be very widely distributed within an area extending ~225 kilometers east-west and 175 kilometers north-south. This is the first region-wide survey undertaken since the 1960s, and it provides an interesting contrast with the conclusions of those earlier surveyors. Lautz (1967), writing several years after *H. irregulare*-caused mortality in shortleaf pine plantations had peaked, implied that the pathogen's niche was limited to young, thinned pine plantations and that as the plantations matured the pathogen's presence and influence were waning. However, although many forest management practices such as thinning and tree harvest do facilitate transmission and development of *Heterobasidion* infestations in forest stands (e.g., Barnard et al. 1991; Woodward et al. 1998), the pathogen is by no means limited to plantations; it also exercises influence over gap creation and forest succession in unmanaged old-growth stands (e.g., Bendel et al. 2006). The current survey revealed the widespread presence of a pathogen that we conjecture played an important role in the Missouri old-growth pine forests of the early twentieth century, that survived the removal of most of these pines by spore dispersal from one isolated pine patch to another, and that continues to cause root rot, mortality, and windthrow on limited scales that match the limited distributions and sizes of pine patches in the Ozarks.

Heterobasidion and landscape structure.

This survey and analysis support the apparent dependence of *H. irregulare* on the presence of shortleaf pine across the Missouri landscape. In the Missouri Ozarks landscape, we did trap *H. irregulare* spores under pure hardwood canopies, but these sites were less than 100 m from shortleaf pine trees. Perhaps because of past logging activity in the Missouri Ozarks, the pine-oak forests of this region constitute a very fine-scale mosaic of patch types where we found it difficult to sample pure hardwood canopies that extend farther than 100m from the nearest pine trees, even though in many cases the pine patch is limited to only one or a few pines. We sampled two sites in hardwood forests just outside the Ozarks range of shortleaf pine (each 36 km distant), plus a much more remote hardwood site (128 km distant), and recovered no spores, despite using large numbers of spore traps per site and despite the presence of the alternative host *Juniperus* on all three sites. The possibility remains that 30 traps per site provides insufficient sensitivity to detect spores in these non-pine sites.

The statistical analysis here did not provide a complete predictive model of pathogen presence and abundance, nor does it support classical hypothesis testing about specific variables. Obviously, a number of other kinds of variables (e.g., topographic, climatic/temperature, and stand history/stand development variables) influence the pathogen's life cycle and epidemiology. Throughout the analyses, distance from the nearest overstory pine consistently (and inversely) influenced pathogen presence, no surprise given previous research on this pathogen's dispersal distances (e.g., Möykkynen et al. 1997). In combination with the failure to recover *H. irregulare* from sites containing redcedar but no pine, we suggest that the pathogen depends on the presence of pine for survival and reproduction across its range in Missouri. Confirming this preliminary conclusion will require more surveys of sites that contain

redcedar and hardwoods but no shortleaf pine at the Ozarks edge.

What we have observed in Missouri forests suggests that *H. irregulare* does not occupy pure hardwood habitats as it apparently does in Italy, where it is non-native and invasive and is increasingly excluding the native *H. annosum*. Gonthier et al. (2012) posed the question whether this ability to occupy oak forests is a novel characteristic in the invaded habitat or also characterizes the pathogen in its native habitat. Our failure to detect this pathogen in pure hardwood forests in Missouri suggests that adaptation to oak habitats may indeed be a novel behavior in Europe, although clearly more surveying is necessary across the pathogen's native habitat in the western, midwestern, and southeastern U.S. (including in Missouri) to establish this.

Landscape-level predictor variables of pathogen presence and abundance other than distance from pine require more exploration. The ratio of pine to hardwood cover in the surrounding landscape, and its spatial configuration, influence the pathogen's presence to at least some degree, as indicated by the consistent presence of patch characteristics in the models. These characteristics, especially the number of pine patches in the surrounding landscape, appear to influence *H. irregulare* presence and abundance through shortening the distance between any given landscape point and the nearest overstory pine (cf. the apparent synergistic interaction between number of pine patches and distance from nearest pine in the models). This has two implications: (1) assessment of Missouri landscapes for likelihood of *Heterobasidion* presence can potentially be done by examining remotely-sensed photos, and (2) a highly fragmented landscape of pine, with many small patches or individual trees, can blanket a landscape with *Heterobasidion* inoculum as efficiently as a landscape with few but large pine patches, if not more so.

Although *Heterobasidion* is not exclusively airborne, the second point is relevant to disease control efforts with pathogens that are. In particular, examining the behavior of this native pathogen relative to host density and configuration can inform efforts to eradicate non-native, invasive, airborne pathogens such as *Phytophthora ramorum*, cause of sudden oak death. *P. ramorum* has proven difficult to eradicate from those landscapes in which eradication efforts have been undertaken (Kanaskie et al. 2009). In some cases, this difficulty is owing to incompleteness of treatment; if any isolated, individual host trees (or regrowing host material) are left at all within treatment areas, they can serve as points of connection to relay airborne inoculum across the landscape, frustrating the treatment goals. The case of *H. irregulare* in the Ozarks stands as an analogue to incomplete treatment, demonstrating the insufficiency of 50-90% host removal to remove an airborne pathogen from a wide landscape over the long term.

Acknowledgements: We thank the following individuals and organizations for assistance, discussion, and permission to survey: LAD Foundation, J. English, J. Greene, J. Hayes, R. Holdo, J. Kabrick, S. McWilliams, J. Mihail, S. Reed, G. Stanosz, S. Wright, Alford Forest and D. Haenke, Mark Twain National Forest, and Missouri Department of Conservation. This work was supported by the University of Missouri Life Sciences Fellowship and a University of Missouri Research Council grant.

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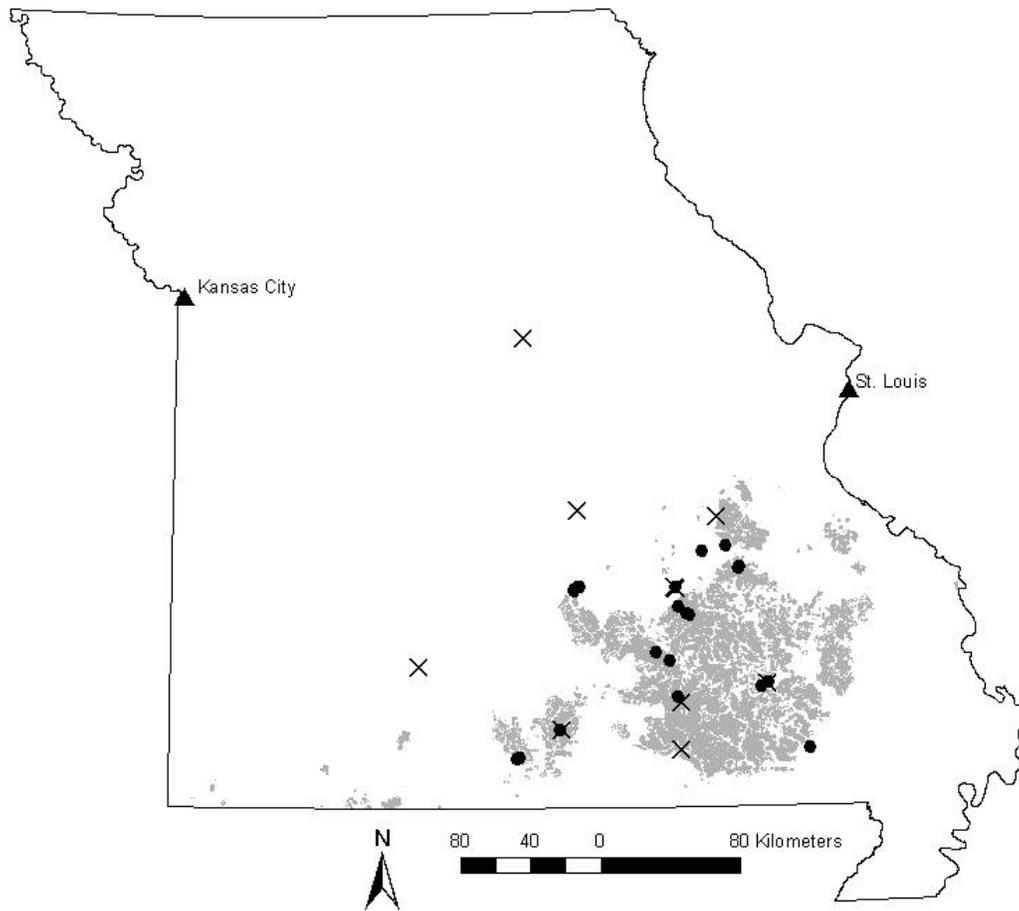


Figure 1. Missouri survey sites for *Heterobasidion irregulare*. A dot denotes a site where we detected the pathogen either as basidiomata or basidiospores; an X denotes a site where we did not detect the pathogen. The shaded area denotes the historic (early 19th c.) native distribution of shortleaf pine in Missouri.



Figure 2. Photos of example surveyed landscapes. A: Big Piney B (BpB) site (mature, planted shortleaf pine stand). B: Fortune Hollow 1 site (pure hardwood stand maintained with periodic burning).

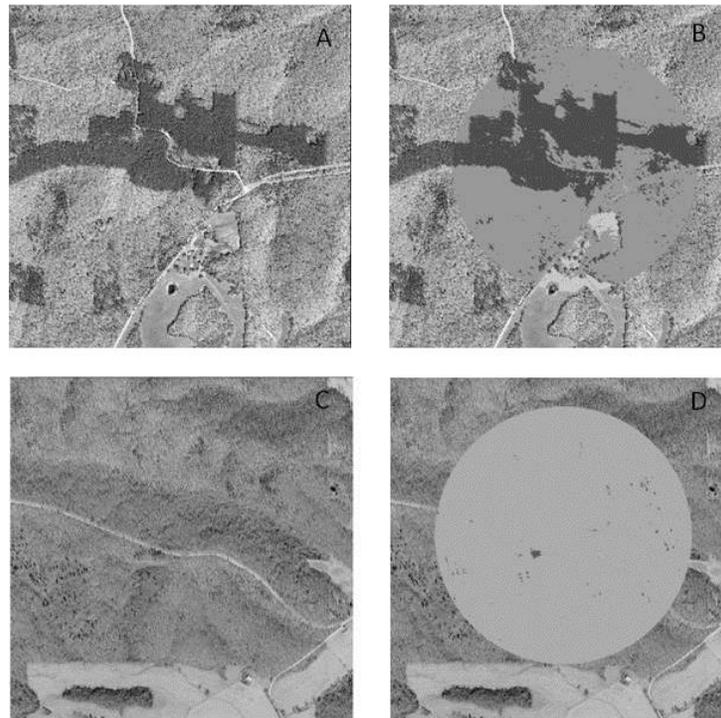


Figure 3. Shortleaf pine distribution in two example Missouri landscapes. A: Leaf-off image of BpB site, showing large, connected patches of shortleaf pine on the landscape. B: BpB site with a GIS-derived supervised classification superimposed on a 500-m-radius circular area around plot center (light gray color denotes hardwood cover). C: Leaf-off image of Clearwater 2 site. D: Clearwater 2 site with 500-m-radius circular supervised classification displaying the patchy, small-scale distribution of pine on the landscape (light gray color denotes hardwood cover).

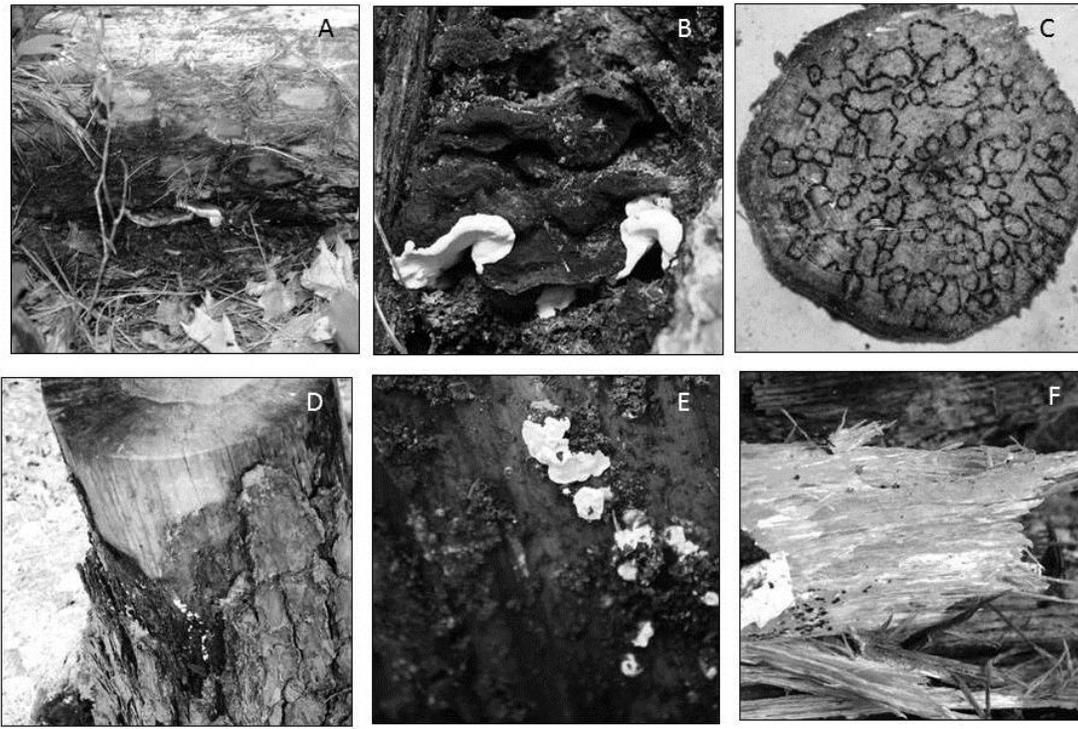


Figure 4. *Heterobasidion irregulare* signs detected at Missouri survey sites. A: Typical basidiome location on underside of fallen pine tree (BpB site). B: Layers of old annual basidiomata on a pine stump with current year's new hymenium on bottom (Alford 2 site). C: Demarcations of individual conidial colonies on shortleaf pine bait disk; this extremely heavy spore load is an outlier in the data set (BpB site). D: Very small basidiomata ("popcorn" or "button" conks) growing beneath bark on old cut stump (Council Bluffs Lake 2 site). E: Close-up of basidiomata in (D). F: Typical white-pocketed, stringy decay caused by *H. irregulare* (Scotia Pond site).

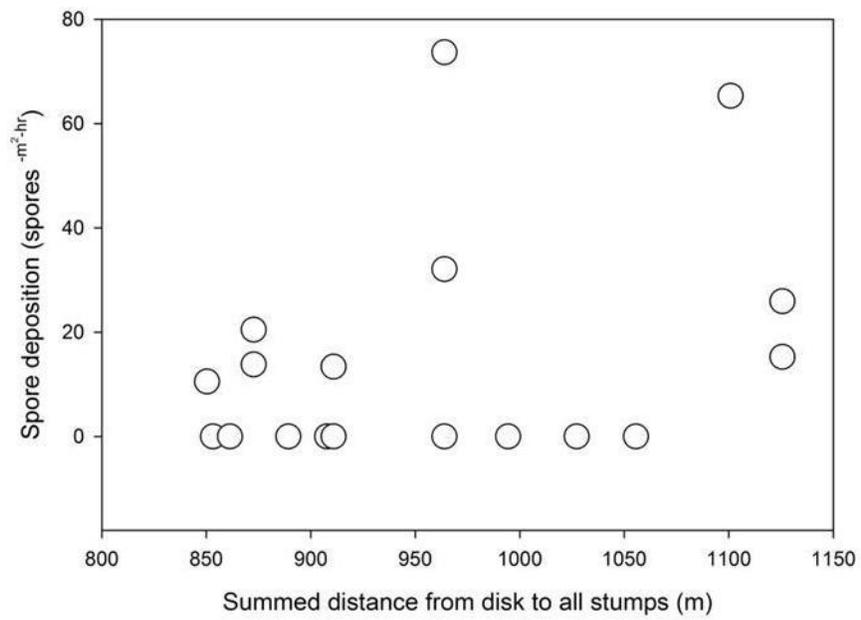


Figure 5. Plot of summed distances between cfu deposition rate at each pine-disk spore trap and all stumps within the plot (Clearwater 2 site).

Table 1. List of sites surveyed with basic characteristics. UTMN = Universal Transverse Mercator “northing” coordinates, Zone 15N; UTME = UTM “easting” coordinates, Zone 15N.

Site	UTMN	UTME	Year	Canopy cover	Pathogen Detection Type		Mean cfu deposition rate (cfu – m ² – hr)
					Basidiomata	Basidiospores	
Alford 1	556843	4068441	2014	pine-oak	X		n/a
Alford 2	555462	4067649	2014	pine-oak	X		n/a
Big Piney B	587881	4163925	2013, 2014	mature pine	X	X	595
Big Piney 1	591040	4166540	2013	hardwood		X	8
Big Piney 2	590211	4165940	2013	young pine		X	14
Big Piney 3	587591	4164475	2013	hardwood		X	89
Clearwater 1	698683	4111932	2013	mature pine	X	X	85
Clearwater 2	695138	4110039	2013, 2014	hardwood		X	8
Clearwater 3	698251	4111620	2013	pine-oak			n/a
Clifty Creek CA	589354	4209856	2014	hardwood			n/a
Compton Hollow CA	499295	4120215	2014	hardwood			n/a
Council Bluffs Lake 1	682401	4178084	2014	hardwood, some pine	X	X	138
Council Bluffs Lake 2	681866	4177210	2014	pine-oak	X		n/a
Fortune Hollow 1	645736	4166313	2013	hardwood		X	4
Fortune Hollow 2	645559	4165610	2014	hardwood			n/a

Fortune Hollow 3	645246	4165836	2014	hardwood			n/a
Greer Crossing	649173	4073371	2014	hardwood			n/a
Harmon Creek	668667	4206399	2014	hardwood			n/a
Hazel Creek	674600	4189641	2014	hardwood, some pine		X	17
Hinkson Creek	558320	4308727	2014	hardwood			n/a
Lewis Lake	649052	4100322	2014	pine-oak			n/a
Noblett Lake 1	580564	4084380	2014	hardwood, some pine			n/a
Noblett Lake 2	580172	4084594	2014	pine-oak	X		n/a
Poplar Bluff	722081	4074740	2014	hardwood, some pine		X	126
Rocky Creek CA	647423	4103656	2014	mature pine	X		n/a
Scotia Pond	647563	4154692	2013, 2014	pine-oak	X		n/a
Red Bluff Campground	660562	4186692	2014	pine-oak	X	X	67
Sinkin 1	653614	4150579	2013, 2014	pine-oak	X	X	107
Sinkin 2	651904	4151008	2013	hardwood, some pine		X	20
Sunklands CA	630530	4129216	2014	hardwood, some pine		X	185
Virgin Pine Demo Area	642643	4124359	2014	mature pine		X	62

Table 2. Candidate models to explain pathogen detection probability (i.e., likelihood of pathogen presence). AICc = Akaike's Information Criterion corrected for small sample size; Δ AICc = change in AICc from best model to other models; w = weighted estimate of relative strength of model (i.e., the probability that this model would be selected as the best one over many repeated runs). Covariate abbreviations: PD = distance to nearest overstory pine; MPS = mean pine patch size within a 500-m-radius circle surrounding plot center; NP = number of pine patches within a 500-m-radius circle surrounding plot center.

Covariates included in model (fixed effects)	AICc	ΔAICc	W
PD	27.55	0.0	0.58
PD, MPS	29.96	2.4	0.17
PD, NP	30.13	2.6	0.16
PD, NP, PD*NP	32.92	5.4	0.04
PD, MPS, NP, MPS*NP, MPS*PD, NP*PD	32.98	5.4	0.04
Intercept only (null model)	35.97	8.42	0.002
NP	38.27	10.7	0.001
MPS, NP	40.97	13.4	<0.001
MPS, NP, MPS*NP	43.94	16.4	<0.001

Table 3. Candidate models to explain spore deposition rate within plots. AICc = Akaike's Information Criterion corrected for small sample size; Δ AICc = change in AICc from best model to other models; w = weighted estimate of relative strength of model (i.e., the probability that this model would be selected as the best one over many repeated runs). Covariate abbreviations: PD = distance to nearest overstory pine; MPS = mean pine patch size within a 500-m-radius circle surrounding plot center; NP = number of pine patches within a 500-m-radius circle surrounding plot center.

Covariates included in model (fixed effects)	AICc	ΔAICc	W
PD, NP, PD*NP	179.47	0.0	0.39
PD	180.21	0.7	0.27
PD, NP	180.35	0.9	0.25
PD, MPS	182.88	3.4	0.07
NP	185.91	6.4	0.02
Intercept only (null model)	188.33	8.4	0.002
MPS, NP	188.38	8.9	0.002
MPS, NP, MPS*NP	190.61	11.1	0.002
PD, MPS, NP, PD*MPS, MPS*NP, PD*NP	201.96	22.5	<0.001