Recovery Plan

for

Red Leaf Blotch of Soybean

caused by

Phoma glycinicola

April 2011

This recovery plan is one of several disease-specific documents produced as part of the National Plant Disease Recovery System (NPDRS) called for in Homeland Security Presidential Directive Number 9 (HSPD-9). The purpose of the NPDRS is to ensure that the tools, infrastructure, communication networks, and capacity required to minimize the impact of high consequence plant disease outbreaks are available so that a reasonable level of crop production is maintained.

Each disease-specific plan is intended to provide a brief primer on the disease, assess the status of critical recovery components, and identify disease management research, extension, and education needs. These documents are not intended to be stand-alone documents that address all of the many and varied aspects of plant disease outbreak and all of the decisions that must be made and actions taken to achieve effective response and recovery. They are, however, documents that will help the USDA to further guide efforts toward plant disease recovery.

Executive Summary

Soybean production in the United States of America (USA) was 2.7 and 3.0 billion bushels in 2007 and 2008, respectively, with a value of nearly 27 billion dollars annually. It is estimated that 3.0 billion bushels of soybean will be produced in 2009. This makes soybean, after corn, the second most valuable crop in the country.

Red leaf blotch (RLB) of soybean is caused by the fungal pathogen *Phoma glycinicola*, formerly known in the plant pathology literature as *Pyrenochaeta glycines*, *Dactuliophora glycines*, and *Dactuliochaeata glycines*. The fungus, although classified as a *Phoma* species, is unique in this genus in that it produces well-defined melanized sclerotia that can either germinate to form infectious mycelia or produce pycnidial structures on the outer surface of sclerotia that produces infectious conidia (spores).

The disease presently occurs in only a few African countries on soybean and a wild legume, *Neonotonia wightii*. Yield losses of up to 50% in soybean were documented in Zambia and Zimbabwe in the 1980s. If the pathogen were introduced into the USA, losses could become substantial; although the pathogen may have limited ability to spread since it does not produce copious amounts of airborne spores like *Phakopsora pachyrhizi*, the causal fungus of soybean rust. Rain-splashed conidia would spread the pathogen causing red leaf blotch with additional dispersal caused by other abiotic factors such as wind, as a contaminant on various tools or clothing and by other biotic factors. The pathogen would most likely survive and over season anywhere in the USA as sclerotia, or possibly pycnidia, in infected plant debris and/or in soil.

Symptoms of RLB on soybean and *N. wightii* include lesions on foliage, petioles, pods, and stems. Lesions expand and coalesce to form large necrotic blotches up to 2 cm in diameter. Heavily diseased plants defoliate and senesce prematurely. Some stages in lesion development resemble lesions caused by other foliar soybean pathogens. Within older lesions, sclerotia develop primarily on the lower surface while pycnidia develop primarily on the upper leaf surface. The fungus is not known to be seedborne, but may be transported along with soil and other debris in grain.

Currently, there is no program monitoring for the introduction of *P. glycinicola* into the USA. Little expertise exists to detect the disease visually and no molecular assays are available. However, the National Plant Diagnostic Network, through its sampling of soybean rust sentinel plots, would be a likely first detector of the pathogen if professionals were trained on the diagnostic symptoms and were able to identify the pathogen. Toward this end, there is a need to develop, provide training materials, and train diagnosticians and growers who may encounter this disease on soybean leaves.

This disease has a relatively low risk of being introduced into the USA because there is little, if any, import of soybean seed and associated debris from infested areas in Africa. Thus, phytosanitary regulations are not needed. If the pathogen were found in the USA, it probably has low potential for rapid spread and could be controlled, but may not be eradicated by fungicides. The most important components of this plan is to identify research needs and to promote education and training so that the disease/pathogen can be identified quickly soon after its arrival

in the USA. Pathogen distribution at the time of detection would affect response recommendations, which may include a quarantine/eradication program at preventing further spread of the pathogen.

Recommended Next Steps

- Develop educational materials and conduct workshops for diagnostians, plant health professionals, and extension agents to raise awareness about the importance of diagnosing RLB and the impact of an introduction of *P. glycinicola* into the USA.
- Develop effective molecular diagnostic techniques to identify *P. glycinicola* from other common foliar soybean pathogens.
- Conduct field experiments where *P. glycinicola* is endemic to evaluate predisposing factors, fungicide efficacy baselines, best practices for application timing, and evaluate soybean resistance and host range of the fungus.
- Develop better prediction models of potential spread of *P. glycinicola* based on distribution of suitable hosts, and more data on the biology and survival of *P. glycinicola*.
- Establish a monitoring system for *P. glycinicola* that uses the current IPM PIPE system.

Red Leaf Blotch of Soybean caused by *Phoma glycinicola*

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I. Introduction

Red leaf blotch (RLB) of soybean is the common name for the disease caused by the fungal pathogen *Phoma glycinicola*. The disease was first reported in Africa in 1957 (Stewart, 1957). RLB occurrence has increased since the 1970s along with soybean production in Africa. RLB is currently a serious threat to production in central and southern Africa with losses of up to 50% reported in countries where it is endemic (Hartman et al., 1987). Other names for RLB are Pyrenochaeta leaf spot, Dactuliophora leaf spot and Pyrenochaeta leaf blotch.

Phoma glycinicola is a culturable, soilborne fungus (Hartman and Sinclair, 1992). It is known to infect soybean (*Glycine max*) and *Neonotonia wightii*, a perennial legume that inhabits the woodlands and grasslands of southern Africa (Leakey, 1964; Stewart, 1957).

The current range of RLB is Cameroon, Ethiopia, Malawi, Nigeria, Rwanda, Uganda, Zaire, Zambia and Zimbabwe. There was one report of the disease outside of Africa, when it was discovered on samples collected in Bolivia in 1982 (Sinclair, 1989). Other reports have not corroborated this find. The disease is not reported in the USA.

The pathogen causing RLB has undergone numerous name changes. Originally, it was named *Pyrenochaeta glycines*, based on the pycnidial stage (Stewart, 1957) and *Dactuliophora glycines*, based on the sclerotial stage (Leakey, 1964). In 1986, both the pycnidial and sclerotial stages were observed in herbarium specimens linking the two epithets to the same fungus (Datnoff et al., 1986). In 1988, a new genus and species, *Dactuliochaeata glycines*, was established to accommodate *P. glycines* and its synanamorph (Hartman and Sinclair, 1988). Most recently, the fungus was classified as a *Phoma* species and re-named *Phoma glycinicola* (Boerema et al., 2004). *P. glycinicola* produces well-defined, melanized sclerotia that on their own can be infectious, or can produce pycnidia on their surface, which then produce infectious conidia (Hartman and Sinclair, 1988). The fungus is unique among the *Phoma* because no other species in that group exhibits such characteristics.

The above cycle, combined with spore survivability and the potential host range will be important factors affecting disease progression should the fungus enter and become established in the USA. In Zambia, up to 19 sclerotia per gram of soil were recovered. Sclerotia kept at 5

°C for 18 months or heat-treated in an oven at 100 °C still germinated at rates of 90% and 22%, respectively (Hartman and Sinclair, 1992) indicating that the sclerotia are very effective survival units. Optimal conidial germination was between 20 and 25 °C, but conidia did not germinate when incubated at 5 ° or 35 °C for over 12 hours (Hartman and Sinclair, 1992). This indicated that these conidia are not long-lived in more extreme temperatures. Leaf disks of ten *Glycine* spp. and six other legumes (cowpea, kudzu, lentil, lima bean, common pea, pigeon pea, and winter vetch) inoculated with *P. glycinicola* became infected (Hartman and Sinclair, 1992). Although soybean and *N. wightii* are the only known natural hosts of *P. glycinicola*, there is a good possibility that more legumes are susceptible. This possibility increases the need for keeping the pathogen out of the USA where other important commercial crops, such as alfalfa, beans and peanuts may be at risk, along with other forage and timber legumes.

II. Symptoms

RLB infection causes similar symptoms on both of its primary hosts, soybean, and *Neonotonia wightii*. Characteristic lesions develop on foliage, petioles, pods, and stems of soybeans. Initial symptoms include lesions that appear first on unifoliolate leaves associated with primary veins (Fig. 1). At this early stage of infection, the disease is easily confused with other diseases like *Alternaria* leaf spot, brown spot, or target spot. Caution is needed before confirming that a plant is infected with the RLB pathogen until the fungal structures form and are identified. As the disease progresses, more characteristic lesions develop on trifoliate leaves, appearing as dark red spots on the upper surfaces and similar reddish brown spots with dark borders on the lower surfaces (Fig. 1). The fungus also causes symptoms on petioles, stems, and pods (Fig. 1).

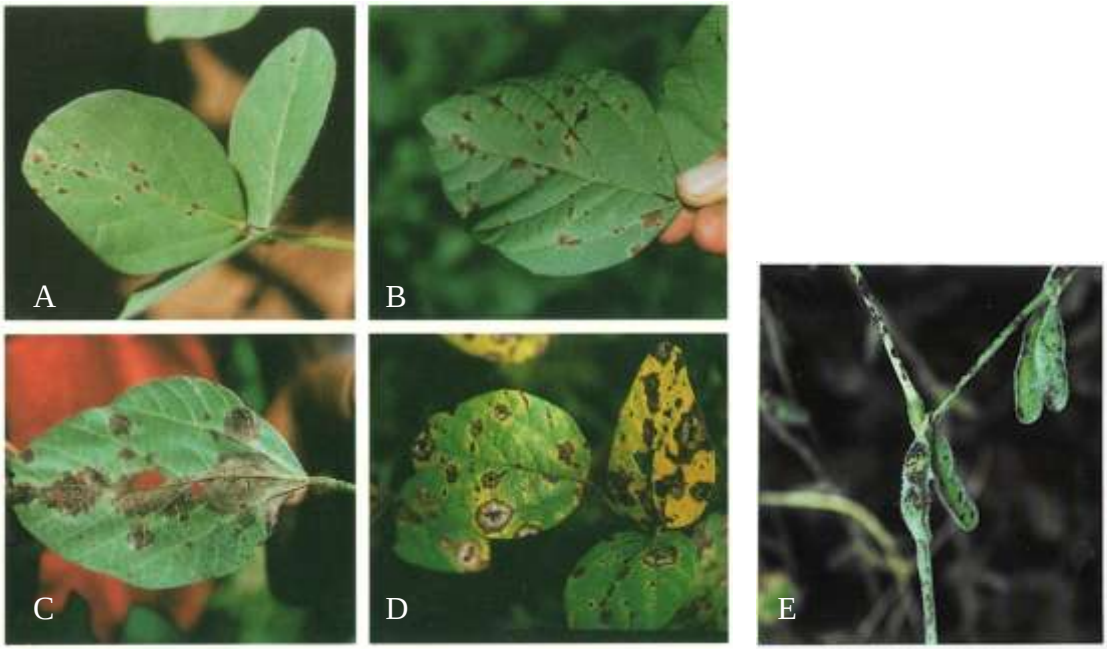

Fig. 1. Red leaf blotch lesions on a young soybean leaf, predominantly along the veins of an upper leaf surface (A). Lesions spreading on a lower leaf surface (B) and coalescing into larger lesions (C). An upper leaf surface showing advanced blotching (D), and lesions on petioles, pods, and stem (E). Reprinted with permission from Hartman et al. (1987).

III. Biology and Spread

The disease cycle was fully characterized on soybean (Fig. 2) (Hartman et al., 1987). Sclerotia reside in the upper soil matrix from decaying leaf litter of either infected soybeans or a primary legume. Infection occurs when rain splashes soilborne sclerotia or conidia from pycnidia onto leaf surfaces, where germination and infection occur (Hartman et al., 1987). Heavily diseased leaves senesce prematurely, and eventually all the foliage from a diseased plant will drop, releasing the sclerotia and pycnidia back into the soil, where they over season and provide the initial inoculum for the next cycle.

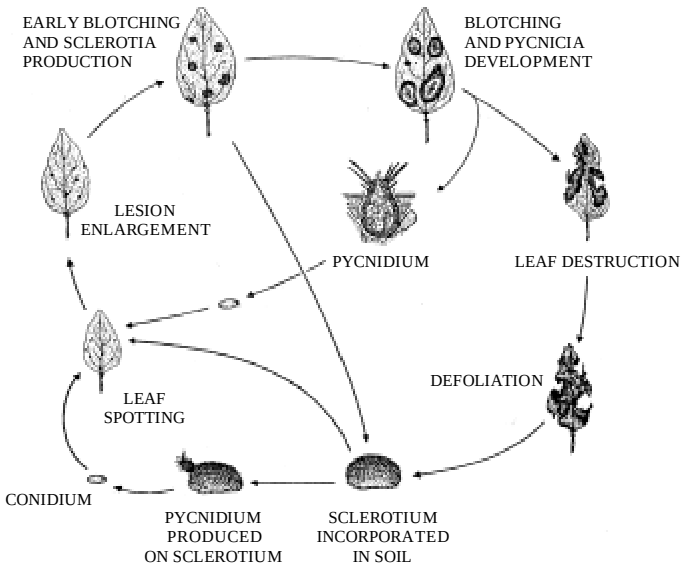

Fig. 2. Red leaf blotch disease cycle. Reprinted with permission from Hartman et al. (1987).

Local spread of the disease occurs when rain showers, water splash, and/or animal or human activities transport the fungal propagules between plants and fields. Sclerotia, residing in the soil, are the primary source of inoculum. There are no studies on transport of sclerotia among fields, but it is reasonable to assume they move similarly to other soil-borne pests, such as soybean cyst nematode, by biotic and abiotic means (Riggs, 2004). Secondary spread via conidia is not well understood (Hartman et al., 1987). The conidia are water-borne and splashed onto leaves in a similar fashion to that found in other *Phoma* species that cause plant diseases (Boerema et al., 2004). Long distance transport of these conidia has not been studied.

There is no evidence that the pathogen is seedborne or airborne. Long distance spread could occur through transport of untreated plant material, via debris accompanying seed from infected fields, or through the movement of contaminated soil.

Pathogen Risk Map. The economic impact of RLB to the USA has not been determined, although a risk assessment has been completed by USDA-APHIS-PPQ-CPHST-PERAL

(Appendix 1). The areas of the USA that have the highest risk of introduction and establishment are the Mississippi River Valley, parts of the eastern Midwest, and the Mid-Atlantic coast (Fig. 3).

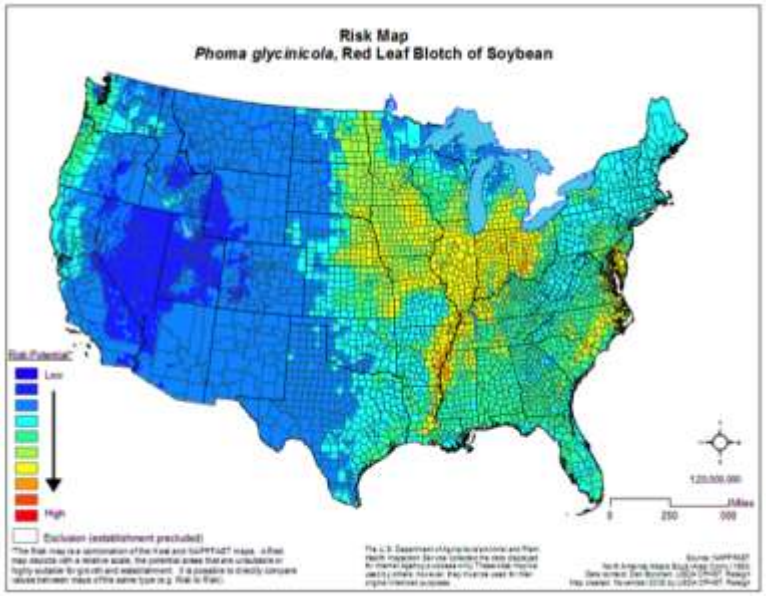

Fig. 3. Risk map for *Phoma glycinicola* introduction and establishment (from Engle and Magarey 2009, by permission).

IV. Monitoring and Detection

Current and Future Surveys**.** There are no surveys monitoring for RLB in the USA and there are no plans for an RLB specific program. It is recommended that the USDA Pest Information Platform for Extension and Education (PIPE), an early warning system to monitor and forecast for the occurrence of soybean rust (SBR), be adapted to include RLB. Soybean leaf samples are collected by a network of scouts and submitted to labs where they are analyzed for the presence of SBR, soybean aphid (SBA) and other diseases. Results are entered in the PIPE database and are then accessible online by the public at http://sbr.ipmpipe.org/cgi-bin/sbr/public.cgi. The online system could be expanded to provide updates and recommendations for RLB should it found in the USA. Both PIPE and NPDN executive committees have been briefed and updated about the Red Leaf Blotch Recovery Plan.

Detection and Diagnosis. Currently, the ability to diagnose this disease is limited to a small group of professionals, and a Cooperative Agricultural Pest Survey is not available to detect it. No methods have been developed to confirm a positive molecular identification of the fungus. Traditional importation of the pathogen into USA plant quarantine facilities to subsequently extract the DNA within the USA inherently increases the pest risk. Therefore, to preclude accidental introductions of RLB into the USA, it will be necessary for several scientists experienced in visual RLB detection to work in partnership with scientists experienced in developing molecular detection methods. Respective experts will identify and collect field samples and process the DNA in-situ. The ribosomal RNA genes of the DNA samples can be

amplified through PCR and sequenced to identify unique regions that can be used to differentiate this pathogen from other fungal pathogens infecting soybean through a Quantitative PCR approach. An international cooperative partnership to identify samples and extract DNA would provide the necessary DNA samples to complete the molecular diagnosis of the fungus.

The NPDN will be crucial in evaluating the spread of this disease in the event this pathogen is discovered in the USA. To fulfill this role, educational materials are needed so participants can recognize the disease from samples that are received in the established system. High quality close-up photos of diseased soybean leaves and microscopic photos of the pathogen are needed for distribution throughout the National Plant Diagnostic Network. A standard operating procedure to aid preliminary identification is being developed, and it is expected to be used before, or as an adjunct to, molecular protocols.

V. Response

Generally, once the detection of a select pathogen is confirmed by a USDA, APHIS, PPQ recognized laboratory, APHIS, in cooperation with the State Department of Agriculture, is responsible for the response. The response is immediate and includes the deployment of teams of experts and survey personnel to the site of the initial detection to conduct investigations and initiate delimiting surveys with recognition of potential movement by contaminated attire. Actions that may be taken include: (1) regulatory measures to quarantine infested or potentially infested production areas to prevent infected material from moving and (2) control measures which may include host removal and destruction, and/or ensuring adherence to required sanitary practices. APHIS imposes quarantines and regulatory requirements to control and prevent both importation and interstate movement of quarantine-significant diseases or regulated articles, and works in conjunction with states to impose these actions in tandem with state regulatory actions that restrict intrastate movement.

After the results of the delimiting survey are known, if the disease is considered generally distributed through commercial and non-commercial plant hosts in an area, options for control will likely include fungicide application. If the disease is isolated, there is a good possibility the pathogen can be eradicated by destroying all infected plants prior to production and deposition of sclerotia. If the detection is late, fields will need to be removed from production indefinitely. Research on sclerotial/pycnidial longevity is needed to address how long a field or location needs to remain in quarantine or under surveillance.

VI. USDA Pathogen Permits

Permit and registration requirements for plant diseases and laboratories are regulated under two authorities: the Plant Protection Act of 2000 (codified at 7 CFR Part 330) and the Agricultural Bioterrorism Protection Act of 2002 (codified at 7 CFR Part 331). Laboratories receiving suspect infected plant material or cultures are required to have PPQ permits. Laboratories possessing, using, or transferring select agents such as *P. glycinicola*, the causal agent of RLB of soybean, are required to be registered; however, diagnostic laboratories that identify select agents or toxins

are exempt from this requirement so long as they complete an APHIS/CDC Form 4 and destroy the culture within 7 days.

The permit requirements of the Plant Protection Act apply to all pests of plants or plant products. This includes importation and interstate movement of pure cultures, arthropod vectors of plant pathogens, diagnostic samples, and infected plant material. The movement of infected plant material, regardless of the pest's quarantine status, requires that the receiving laboratory have a permit. The importation and/or interstate movement of soil is similarly regulated when the intent is to isolate microbes that may be pests of plants or plant products. For guidance on the permitting of plant pests and soil samples, consult the PPQ permit website at: http://www.aphis.usda.gov/ppq/permits/ or contact PPQ Permit Services Customer Services at (301) 734-0841.

The Agricultural Bioterrorism Protection Act specifies requirements for possession, use, and transfer of organisms listed as select agents or toxins, such as the newly listed *P. glycinicola*. Once an unregistered diagnostic laboratory identifies a presumptive select agent or toxin, they must immediately notify the Agriculture Select Agent Program (ASAP), complete an APHIS/CDC Form 4 within 7 days, and either destroy or transfer the agent to a registered entity within 7 days (prior approval of the ASAP required). If a diagnostic laboratory held part of a screened sample (or culture) for voucher purposes, and the sample forwarded to the USDA Beltsville Laboratory was identified as positive for a select agent or toxin, then the USDA Beltsville Laboratory will notify immediately both the ASAP and the sending diagnostic laboratory that a select agent or toxin has been identified. The USDA Beltsville Laboratory will submit the APHIS/CDC Form 4 within seven days, and all unregistered labs will either destroy or transfer the samples to a registered entity within 7 days of receipt of the results. Agriculture Select Agent Program personnel must have an opportunity to witness the destruction of the sample(s) or culture(s) within that time period. Clarification of these requirements and other information related to adherence to the select agent regulations is available at: http://www.aphis.usda.gov/programs/ag_selectagent/ and http://www.selectagents.gov, or call (301) 734-5960, Agriculture Select Agent Program.

VII. Economic Impact and Compensation

Yield losses of up to 50% were reported in Zambia and Zimbabwe (Datnoff et al., 1987; Hartman and Sinclair, 1996). Statistics from other affected areas are not available.

Soybean production in the USA was 2.7 and 3.0 billion bushels in 2007 and 2008, respectively, and valued at nearly 27 billion dollars annually. It is estimated that 3.0 billion bushels will be produced in 2009. This makes soybean, after corn, the second most valuable crop in the USA (http://www.ers.usda.gov/News/soybeancoverage.htm).

Table 1. Soybean production for the top five states in the USA in 2008[1]

State	Production [2]	Value[3]
Iowa	444,820	4,292,513
Illinois	427,700	3,998,995

Minnesota	264,100	2,535,360
Indiana	244,350	2,272,455
Nebraska	225,990	2,124,306

[1](http://www.nass.usda.gov/QuickStates/index2.jsp)
[1]Thousand bushels
[3]Thousand dollars

Currently, countries that are most affected by RLB are in hot, humid regions. While the top five USA soybean-growing states are temperate, and the lesser regions of production in the southern states are sub-tropical, this disease could be a threat to soybean production anywhere in the USA. Although the behavior of the pathogen in temperate climates has not been previously studied, it seems likely that the pathogen could survive in the temperate climate of the USA. In this way, this fungus may exhibit similarities to another soybean disease caused by *Sclerotinia sclerotiorum* where sclerotia readily survive in USA temperate climates (Grau and Hartman, 1999). Unfortunately, direct comparisons between these sclerotia-producing fungi are not available in the scientific literature.

Compensation by the Risk Management Agency (RMA) for a loss caused by RLB is available, provided the producer can verify that available control measures were applied. If there are no effective control measures available or there are insufficient amounts of chemicals available for effective control, resulting loss of production would be covered. It will not be a covered loss if there are sufficient control measures available, but the insured elects not to use them because he/she feels the cost is too high.

VIII. Mitigation and Disease Management

Any disease mitigation strategy that is utilized must be coordinated among Federal, State and local regulatory officials because efforts by individuals are not expected to provide widespread disease control.

Prevention/Exclusion. RLB is not yet reported in the USA and has a relatively low risk of being introduced into the USA because there is little, if any, import of soybean seed and associated debris from infested areas in Africa. Thus, phytosanitary regulations are not needed. If the pathogen were found in the USA, it probably has low potential for rapid spread and could be controlled, but probably not eradicated, by fungicides. Since *P. glycinicola* was recently added to the Agriculture Select Agent Program, reflecting heightened concern over the threat posed by the disease to American agriculture, continued exclusion of this disease through port activities is an essential initial step in the mitigation and disease management strategy.

Epidemiological Studies. Yield loss studies have been completed (Hartman and Sinclair, 1996); however, there are other factors that are not well understood. This includes the predisposition of plants to the fungus, and the spread of the various propagules (conidia, pycnidia, and sclerotia) by water, wind, and other means. Disease gradients, to measure short and long distance spread, are not known, but would be useful in modeling the spread of RLB.

Germplasm. In 1982 and 1984, all USA-grown commercial cultivars were susceptible to this disease in field tests in Zambia and Zimbabwe (Sinclair, 1989) and in trials conducted before 1992 based on Hartwig's southern USA soybean germplasm collection – approx. 5000 lines that were tested in Zimbabwe (pers. comm., Clive Levy). Host resistance may be a viable option in managing RLB if this disease enters and persists in the USA. Additional germplasm evaluations are needed to discover good sources of resistance.

Biological and Cultural Control. There are no biological controls available at the present time. Information on the role of cultural practices in suppression of *P. glycinicola* is not available in the scientific literature. Whether cultural practices effective against other soybean diseases will be effective against RLB is not known.

Chemical Control. Recommendations include the application of fungicides. The fungicide fentin actetate was effective for control of red leaf blotch in Africa, but this fungicide is no longer in production. There is no information available to suggest whether market forces would favor the development of a novel fungicide for this disease, or encourage companies to seek an additional registration for an existing compound. Current registered fungicides in the USA need to be tested for the efficacy to RLB.

Eradication. Early detection and the destruction of infected material may make eradication possible if there is a single entry point with limited spread. This may be followed with planting of non-host crops, like corn, rice or wheat, and extensive monitoring for the reasonable future to check for recurrence of the disease. Eradication may not work if multiple entries have occurred or if the spread is extensive beyond a single entry point.

Education. Education efforts about RLB at all levels are needed. Importers, growers, diagnosticians, and cooperative extension experts must become aware of this disease and trained in its identification. Information on how to distinguish this disease from others that resemble it (Hartman et al. 1999) will be critical for detection at ports of entry and in the field. These latter entities are instrumental in disseminating additional information about the threat posed by this disease, and any effective cultural and chemical practices designed to mitigate its spread that are developed and deployed. Early detection relies on accurate information, and early detection is absolutely necessary to provide the time needed for successful eradication efforts if this disease enters and becomes established in a localized area. For this to be accomplished, growers and diagnosticians need an awareness of the disease and most have the ability to identify it. Should there be a positive identification, cooperative extension experts can play a role in further educating growers about the threat posed by the disease and the cultural and chemical practices that mitigate its spread needs to intensify. Specifically, educational materials explaining how to distinguish this disease from others that resemble it need to be prepared and distributed (Hartman et al. 1999).

Recommended disease management strategy. The recommended disease management strategy will depend upon the severity of the first detection and the spatial distribution and the severity associated with the pathogen. If local in occurrence, the destruction of infected material may make eradication a possibility if there is a single entry point with limited spread. Subsequent planting of non-host crops, like corn or rice, and/or ongoing monitoring is needed to preclude

disease reoccurrence. Conversely, if the distribution extends beyond a point source (e.g. multiple fields or states), then disease management will rely on emergency-labeled fungicides. While additional registrations may fill some gaps and allow a crop to be produced, the costs associated with crop production will increase. Further, growth of a susceptible crop will require disease monitoring followed by implementation of a fungicidal plan immediately after positive confirmation of the disease. Over time, long term studies evaluating other aspects of disease suppression and management (e.g. tillage and rotation studies) will be as essential as the discovery and development of sources of host resistance.

IX. Infrastructure and Experts

A research project concerning RLB is active at the USDA/ARS Bio-Safety Level 3 Plant Pathogen Containment facilities in Ft. Detrick, MD. (http://www.ars.usda.gov/research/projects/projects.htm?ACCN_NO=411489). The title of this research project is "Identification, characterization, and biology of emerging foreign fungal plant pathogens" (Project Number: 1920-22000-035-00). The focus of this project, primarily implemented by Paul Tooley, is to culture and maintain the fungus, develop inoculation techniques, and evaluate host resistance.

The following scientists have experience working with RLB or *P. glycinicola* and can be contacted specifically for the expertise listed:

Glen Hartman, USDA-ARS, Urbana, IL
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Expertise: Field diagnosis, epidemiology, and disease management; fungal biology and taxonomy.

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Expertise: Field diagnosis, epidemiology, and disease management; fungal biology and taxonomy.

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Expertise: Field diagnosis, epidemiology, and disease management.

Paul Tooley, USDA-ARS, Ft. Detrick, MD
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Expertise: Fungal biology, host resistance.

Lawrence Datnoff, Louisiana State University, Baton Rouge, LA
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Expertise: Field diagnosis, epidemiology, and disease management; fungal biology and taxonomy.

X. Research, Extension, and Education Priorities

Research Priorities (high/low priority; long/short term)

- Determine the overwintering potential in USA simulated conditions to better understand the threat posed by the pathogen/disease. (high priority; short term)
- Evaluate efficacy and application methods of fungicides currently registered for use on soybean. (high priority; short term)
- Develop effective molecular diagnostic techniques to identify *P. glycinicola* from other common foliar soybean pathogens. (high priority; short term)
- Determine effective measures to control, confine, or destroy the pathogen in the field through collaboration with scientists in countries with RLB. (high priority; long term)
- Develop more epidemiology studies to determine predisposition factors involved in disease development and to evaluate disease gradients to measure short and long-term spread. (high priority; short term)
- Discover sources of resistance, understand disease resistance mechanisms, and develop resistant commercial varieties. (high priority; long term)
- Develop better prediction models of potential spread of *P. glycinicola* based on knowing more about alternative hosts in the U.S. that might serve as sources of inoculum of *P. glycinicola*. This would include host range studies in RLB endemic areas (low priority; long term)
- Evaluate the efficacy of various biocontrol agents that are commercialized for use on other soybean diseases like Sclerotinia stem rot for the control of RLB. (low priority; long term).

Extension Priorities (high/low priority; long/short term)

- Provide outreach materials for diagnostians, soybean growers, crop advisors, and industry on RLB diagnosis and disease management (high priority; short term)
- Disseminate information gained through research to other researchers at meetings, workshops, and other venues. (high priority; short term)
- Establish a field monitoring system compatible with current programs like the IPM PIPE. (high priority; long term)
- Distribute fungicide guidelines. (low priority; long term)

Education Priorities (high/low priority; long/short term)

- Develop training materials on detection, monitoring, and management of RLB. (high priority; short term)
- Host a diagnostician workshop in Beltsville for provisional diagnostic standard operating procedure development and training (cultural characteristics, digital diagnostic refresher, PCR). (high priority; short term)
- Develop training materials for port of entry Safeguarding Specialists (USDA) and Department of Homeland Security personnel. (high priority; short term)
- Conduct first detector training – laboratory (NPDN) and field (PIPE). (high priority; short term)

XI. Timeline for Recovery

Recovery time will depend on the location and intensity of the infestation. For example, if only a plot in a research field or a small area in a grower's field is discovered, then recovery may essentially be one year after treatment. The infested area and the immediate surrounding area (up to 100 meters) would be isolated for treatment. Treatment would include a fungicide (Section 18 Emergency Exemption) and a soil fumigant. The area would be planted to soybeans in continuous years and monitored for the presence of RLB. If it occurs during the second season, treatments would have to be repeated. In this case, eradication would be the goal.

If the area of infestation were greater than a local infestation as described above, then recovery time would be expected to be much greater. Eradication of the disease would not be possible and management options would be considered that would include tactics of fungicide use and host plant resistance. Both options may be effective management tools, however, fungicides would be the first line of management (used the first year and subsequent years) and host resistance would follow (it could be up to 10 years before resistance varieties would be available in the market).

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Web Resources

National Laboratory for Soybean Disease Research: http://www.soydiseases.illinois.edu/

National Plant Diagnostic Network: http://www.acronymfinder.com/National-Plant-Diagnostic-Network-%28NPDN%29.html

Appendix 1. Risk Assessment for Red Leaf Blotch

WEATHER-BASED PEST RISK MAPPING PROJECT

RISK ASSESSMENT:

Phoma glycinicola, Red Leaf Blotch of Soybean

Cooperative agreement between NCSU and

USDA-APHIS-PPQ-CPHST-PERAL

I. Rationale

We developed this risk assessment to assess the climatic favorability of the United States for *Phoma glycinicola*.

Photo: Rikus Kloppers/PANNAR Research, Greytown, South Africa.

II. Life History and Biology

Red leaf blotch (RLB) of soybean (a.k.a. Pyrenochaeta leaf spot, Dactuliophora leaf spot, and Pyrenochaeta leaf blotch) is caused by *Phoma glycinicola* Gruyter & Boerema, formally known as *Pyrenochaeta glycines* Stewart, *Dactuliophora glycines* Leakey, and *Dactuliochaeata*

glycines Stewart. Observed hosts of RLB are *Glycine* spp., *Neonotonia wightii, Pisum* spp*., Phaseolus* spp., *Vigna unguiculata, Pueraria lobata, Cajanus cajan*, and *Vicia villosa* (Hartman and Sinclair, 1992), potentially indicating that most legumes, including alfalfa (*Medicago* spp.), could be hosts. Yield losses on soybeans of up to 50% have been reported in Zambia and Zimbabwe (Hartman et al., 1987).

The disease cycle has been fully characterized on soybean (Hartman et al., 1987). The source of the primary inoculum is from sclerotia residing in soil as this fungus is not seed borne. The secondary spread, via pynciopsores, is not documented but presumably the spores are splash borne like they are with other Phoma species (Boerema et al., 2004). Initial RLB symptoms are lesions associated with primary leaf veins that appear on unifoliolate leaves. During the early stages of infection the disease may be confused with Alternaria leaf spot, brown spot, and target spot. More characteristic lesions, dark red spots on the upper leaf surface with similar reddish brown and dark bordered spots on the lower leaf surface, develop on trifoliate leaves. Symptoms occur not only on the leaves, but also the stems, petioles, and pods. Similar to most fungal soybean diseases, wet and humid conditions promote RLB disease development (Hartman and Sinclair, 1992). Lesions expand and coalesce to form large necrotic blotches up to 2 cm in diameter with heavily diseased plants defoliating and senesce prematurely. Within older lesions, sclerotia develop primarily on the lower surface of leaves; pycnidia develop primarily on the upper surface. Movement in the canopy is through rain splash while long distance movement is accomplished through transport of untreated plant material, through infected debris that accompanies seed, or through movement of inoculated soil on machinery (Hartman and Haudenshield, 2008).

Control of RLB in Africa, where it is endemic, is through fungicide application. Currently, these fungicides have not been labeled for RLB treatment in the US. There is no known biological for RLB. A more cost effective method of control is host resistance. Unfortunately, to date there are no US cultivars that have resistance that have been reported. The best defense for RLB is exclusion from the US production zones.

III. Prediction Model

Host density

We created a risk map based on percentage host acres per county from National Agricultural Statistics Service data (http://www.nass.usda.gov/). We calculated the density by dividing the total number of acres in *Glycine* spp., *Phaseolus* spp., and *Pisum* spp. by the total acres per county (Fig. 1).

Prediction model

We created a simple prediction model based on the average history from the last ten years of favorable days and defined a favorable day as having a minimum temperature above 5°C, an optimum temperature of 22.5°C, a maximum temperature below 35°C, at least 0.5 hours of leaf wetness, and 2mm of precipitation (Hartman and Sinclair, 1992). The average history of favorable days were grouped into ten classes: 0, 0-2, 2-15, 15-30, 30-45, 45-60, 60-75, 75-90, 90-120, 120+ days (Fig. 2). Risk maps were created with the NCSU APHIS Pest Forecasting System (NAPPFAST) system (Magarey et al., 2007). The NAPPFAST system uses a web-based graphical user interface to link climatic and geographic databases with templates for biological modeling. The NAPPFAST system includes two daily weather databases with over 30 years of records. The global database is based upon the National Centers for Environmental Prediction (NOAA/NCEP) Global Reanalysis II data set (Kalnay et al., 1996). This data set is a numerical grid created for use as input data for meteorological models. The spatial resolution of the grid is 32 km, which has been resampled from a 1.875 degree (210 km) resolution. Station data from the International Station Hourly (ISH) data (Lott et al., 2001) were used to supplement the NCEP backbone. The North American database includes over 2000 stations for North America (Magarey et al., 2007). The input weather data was interpolated to a 10 km^2 resolution using a 3-D multivariate interpolation (Splitt and Horrel, 1998).

IV. Results and Discussion

The majority of soybean, bean, and pea production is located in temperate regions of the US (Fig. 1). While alfalfa production was not included in the host map, it would also mostly fall in the temperate regions and the arid Pacific Northwest of the US. These areas are not conducive for rapid establishment and proliferation of *P. glycinicola* according to the NAPPFAST ten year average history model (Fig. 2). Unfortunately, the overwintering sclerotia would allow establishment of *P. glycinicola* in the temperate areas over time. These localized sclerotia would then be able to cause epidemics in years when the environment does not follow the ten

year average history. The areas of the US that do have the highest risk of introduction and establishment are the Mississippi River Valley and the Mid-Atlantic coast (Fig. 3) when considering the US susceptible crop production and NAPPFAST model. It is the authors' opinion that the infection of kudzu (*Pueraria lobata*) would allow for rapid establishment and production of copious amounts of *P. glycinicola* sclerotia in the soil in the Southeast US. Unfortunately, there is no documentation of the number of acres per county of kudzu, therefore it could not be incorporated into the model. Soybean and other susceptible hosts production in areas with both kudzu and *P. glycinicola* populations could potentially become economically unfeasible.

V. Authors

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VII. Figures

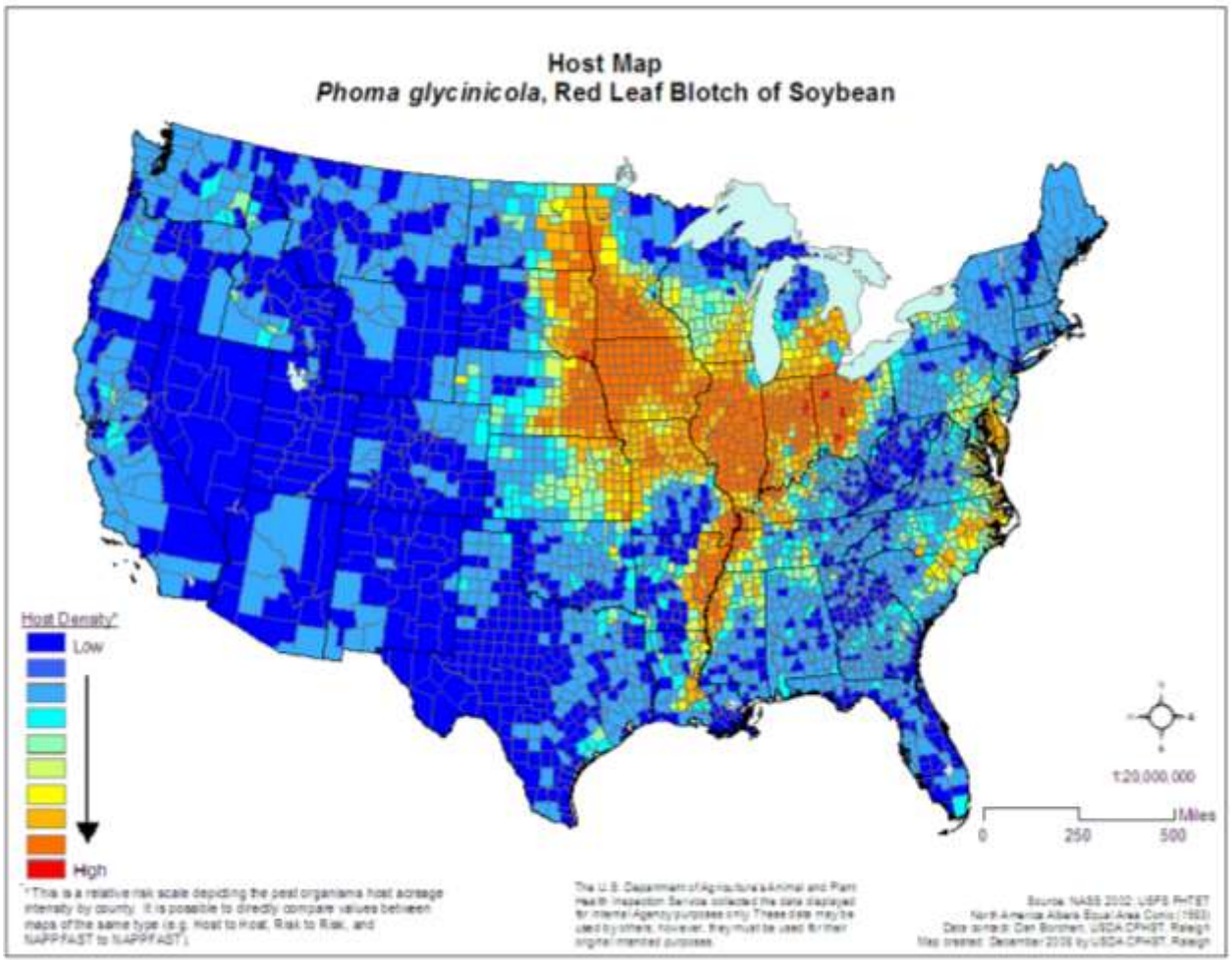

Figure 1. Percentage of county acres in *Phoma glycinicola* susceptible host production in the United States.

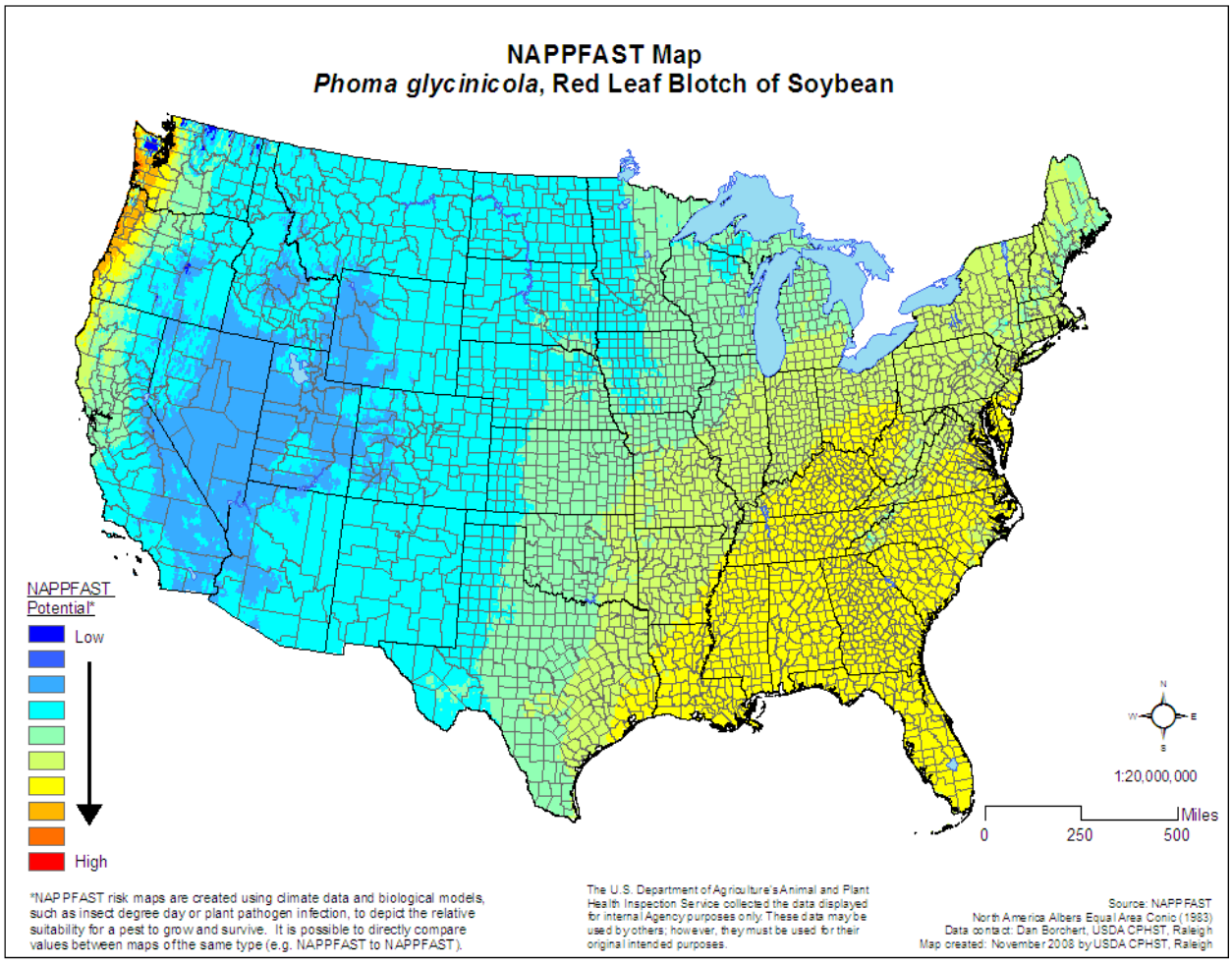

Figure 2. NAPPFAST prediction model map for favorable days for *Phoma glycinicola* infection.

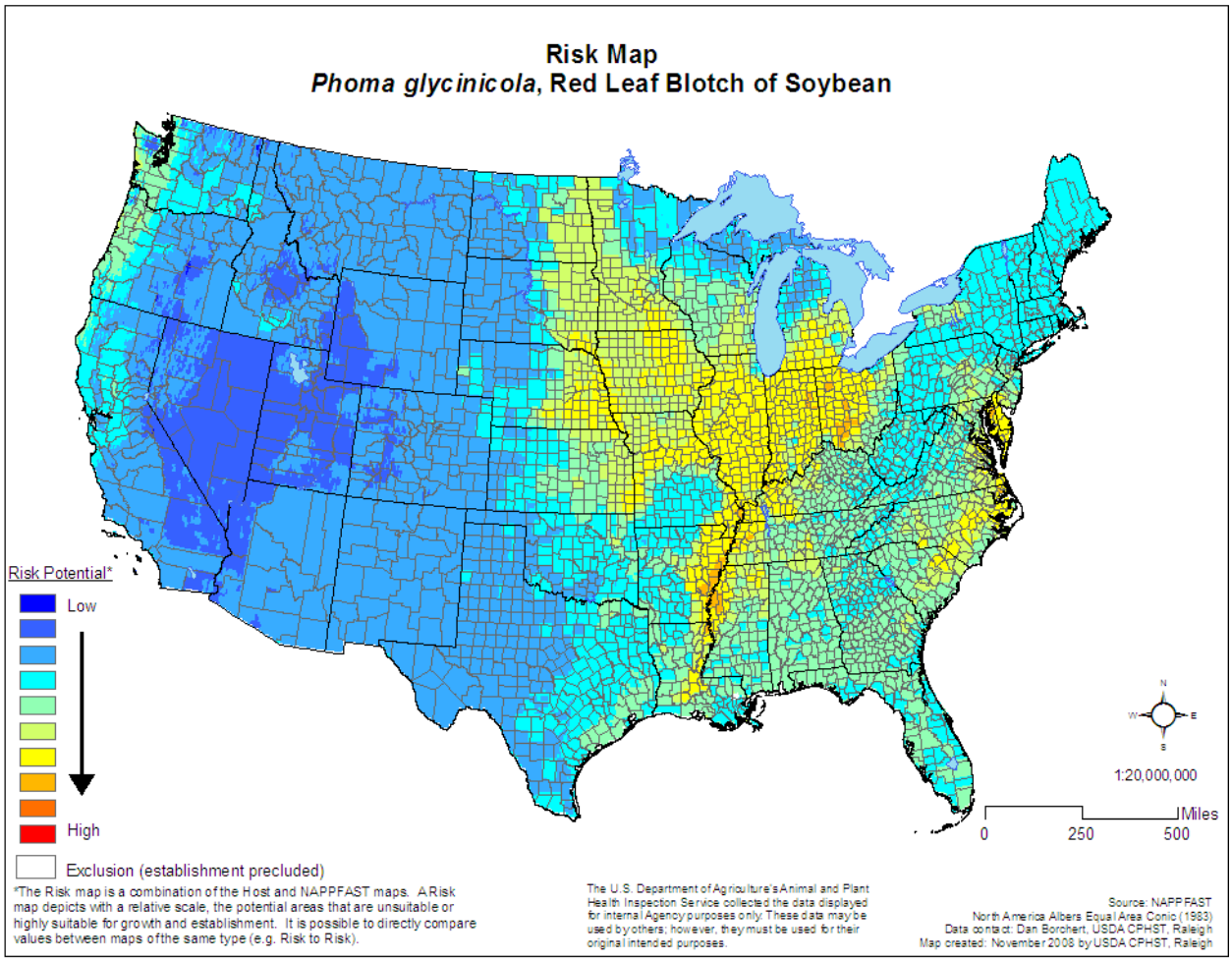

Figure 3. Final risk map for *Phoma glycinicola* introduction and establishment.