Status of Scientific Evidence on Risks Associated with the Introduction into the Continental United States of *Phakopsora pachyrhizi* With Imported Soybean Grain, Seed and Meal

United States Department of Agriculture
Animal and Plant Health Inspection Service
Plant Protection and Quarantine

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EXECUTIVE SUMMARY

Soybean rust is caused by either of two fungal species, *Phakopsora pachyrhizi*, or *Phakopsora meibomiae*. The Asian strain of soybean rust (SBR) caused by *P. pachyrhizi* is extremely aggressive and is listed as a select biological agent (Title 7, Code of Federal Regulations, Part 331.2). *P. meibomiae* is less aggressive on soybean, and is mostly found in the Tropical Western Hemisphere. Soybean rusts caused by these two species have been reported in most soybean producing areas of the world, except North America and Europe.

Recent outbreaks of the Asian strain of soybean rust (SBR) have occurred in South America causing United States soybean producers to ask APHIS to re-evaluate the entry status of soybean grain, seed and meal from countries where SBR is known to occur. This document describes the current information available to APHIS. It will be used to inform the development of a risk assessment.

Based on this information, the following main conclusions have been drawn: As a result of commercial production and trade of soybean seed, grain and meal and the biological characteristics of SBR:

- Clean soybean seed, clean soybean grain and soybean meal are not pathways for the introduction of SBR.
- *Phakopsora pachyrhizi* is an obligate parasite and its spores rapidly lose viability after the plant dies. It will not infect or colonize dead or dried plant tissue.
- Since becoming established in South America a few years ago, SBR has spread rapidly and is expected to continue spreading naturally in the Western Hemisphere and eventually to the United States.
- SBR introduction into the United States could cause significant crop losses, ultimately resulting in widespread and complex market disruptions.
- Soybean leaf debris associated with "foreign material' found in soybean grain presents a theoretical pathway for the introduction of SBR. However, normal commercial practices minimize the presence of "foreign material" to less than 2%. Moreover, as it is normal commercial practice to harvest soybeans after the plants have been defoliated, leaf debris should compose only a fraction of the "foreign material"; therefore, making "foreign material" found in soybean grain an unlikely pathway for the introduction of SBR.

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INTRODUCTION

Soybean rust can be caused by either of two fungal species from the genus *Phakopsora*. *Phakopsora pachyrhizi* Sydow, is the fungal species known as the Asian, Australian, or Old World rust strain. *Phakopsora meibomiae* (Arthur) Arthur, also causes soybean rust and is referred to as the tropical, Latin American or New World rust strain. This document focuses only on soybean rust (SBR) caused by *P. pachyrhizi*..

Phakopsora pachyrhizi is an air-borne fungal pathogen that is not present in the continental United States. Under conducive environmental conditions, this pathogen could cause serious economic and crop losses in major soybean production regions of the United States. The probability of long-distance spread of *P. pachyrhizi* across U.S. borders may be reduced through strong international cooperation to reduce inoculum levels beyond our borders. However, it is anticipated that the disease will eventually reach the U.S. and establish in major soybean growing regions via wind currents. The confirmation of the presence of Asian SBR in soybean production regions of Argentina, Bolivia, Brazil and Paraguay has lead to heightened concerns regarding the potential for spread to the continental United States. In particular, there is concern about potential introduction of Asian SBR in the importation of soybean seed, meal, and grain of host plant members of the pea and bean family (Leguminosae).

The Center for Plant Health Science and Technology (CPHST), Plant Protection and Quarantine (PPQ), Animal and Plant Health Inspection Service (APHIS) of the United States Department of Agriculture (USDA) has compiled the following available information regarding potential pathways for the introduction of *Phakopsora pachyrhizi* into the United States.

BACKGROUND

History and Geographic Distribution

Formal distinction between the two fungal species causing soybean rust did not occur until 1992 (Ono *et al.*), so earlier references inadvertently include the distribution of *P. meibomiae* along with that of *P. pachyrhizi.* Global distribution of both fungal pathogens is provided in Figure 1. *P. pachyrhizi* is the cause of the most recent and yield limiting outbreaks of SBR in South Africa and South America. This document focuses on *P. pachyrhizi*, which is considered to be the more virulent fungal pathogen.

Soybean rust caused by *Phakopsora pachyrhizi* was first observed in Japan in 1902, and by 1934 the pathogen was found throughout most Asian countries and in Australia (Bromfield, 1984). The current distribution of *P. pachyrhizi* includes countries in Africa, Asia, Australia and most recently South America (Soybean Rust Meeting, June 26-27, 2002). Asian SBR causes serious crop losses in most infested soybean production regions. The disease has not yet been detected in North American or Europe.

SBR has been reported in the main soybean growing regions of Brazil including the states of Parana, Rio Grande do Sul, Mato Grosso do Sul and Goais. About 2.6 percent of the 15 million hectares planted to soybean in Brazil tested positive for the *P. pachyrhizi*, and the disease caused 10 percent loss in the 2000-2001 soybean crop in affected fields (e-mail from John C. Baize, United Soybean Board Marketing, Trade Analysis and Audit and Evaluation Committee). In the Brazilian regions of Mato Grosso do Sul and Goais, SBR reduced yield by up to 75% in some fields (email from John C. Baize). During 2001-2002 growing season, Brazil growers lost approximately 112 thousand tons of their annual soybean production (Soybean Rust Meeting, June 26-27, 2002). *P. pachyrhizi* is suspected to have been introduced to Brazil via air currents from Africa and/or Asia. Asian SBR is currently causing yield losses of up to 70 percent in countries such as Zimbabwe and Taiwan (email from John C. Baize).

Phakopsora pachyrhizi is present throughout the soybean production areas of Australia, Burma, Cambodia, China, Congo, Ghana, India, Indonesia, Japan, Kenya, Korea, Malaysia, Mozambique, Nepal, Nigeria, New Guinea, Philippines, Rwanda, Sierra Leone, South Africa, Taiwan, Thailand, Uganda, Vietnam, Zambia, and Zimbabwe (Bromfield, 1984; Ono *et al.*, 1992; Pretorius *et al.*, 2001; NPAG, 2002; Miles *et al.*, 2003). Soybean rust was observed in Hawaii in 1994 on the islands of Oahu, Kakaha, Kauai, and Hilo (Killgore *et al.*, 1994), where it is thought to have arrived in fresh soybean plants smuggled in for use in ethnic Laotian cuisine (Tschanz, personal communication). *P. pachyrhizi* was first detected on the South American continent in Paraguay in 2001, where it was widely spread, then was wind spread across the border into Argentina. Between 2001-2003, it became established and widespread in soybean production regions of Brazil (Rossi, 2003). SBR was recently found in Bolivia in July, 2003 (personal communication, J. T. Yorinori, Embrapa Soja, Brazil).

Crop loss estimates for soybean rust due to *Phakopsora pachyrhizi* range from 10-80% (Bromfield *et al.*, 1980; Casey, 1981; Dowler and Bromfield, 1983; Manandhar and Joshi, 1983; Bromfield, 1984; Wrather *et al.*, 1997; Sinclair and Hartman, 1999; Kawuki *et al.*, 2003). According to Clive Levy, 2003 (personal communication), in Zimbabwe losses due to *P. pachyrhizi* can reach up to a 100% under disease conducive field conditions.

Biology

Taxonomic position

Phylum:	Basidiomycota
Class:	Urediniomycetes
Order:	Uredinales
Family:	Melampsoraceae

Synonyms: *Phakopsora sojae* Fujikuro
Phakopsora calothea H. Sydow
Malupa sojae (P. Hennings) Ono, Buritica, & Hennen comb. nov. (Anamorph)
Uredo sojae P. Hennings
(For additional synonyms, see Green, 1984; and Ono *et al*., 1992)

Teleomorph: *Phakopsora pachyrhizii* H. Sydow & Sydow: telia are crustose, 2~ to 7~ spores layered, chestnut-brown to chocolate-brown, and subepidermal.

Anamorph: *Malupa sojae* (P. Hennings) Ono, Buritica & Hennen. comb. nov.: uredinial sori minute, scattered or in groups on discolored lesions, subepidermal in origin; urediniospores are obovoid to broadly ellipsoid, 18-38 x 13-29 µm, germ pores on an equatorial zone or scattered and the walls are densely echinulate, colorless to pale yellowish brown.

Common names: Soybean rust, soybean rust fungus, rust of soybean, Asian soybean rust, Old World soybean rust (CABI, 2002).

Genome: The *P. pachyrhizi* genome is currently being sequenced (Miles *et al.*, 2003).

According to Alexopoulos, *et al.* (1996) a rust fungus may produce as many as five different stages in its life cycle:

♦ stage 0	Spermagonia bearing spermatia (n) and receptive hyphae (n)
♦ stage I	Aecia, bearing aeciospores (n + n)
♦ stage II	Uredinia bearing urediniospores (n + n)
♦ stage III	Telia bearing teliopores (n + n → 2n)
♦ stage IV	Basidia bearing basidiospores (n).

Phakopsora pachyrhizi is usually described from the uredinial and telial stages; production of all five stages is uncertain (Green, 1984). Like all rust fungi, *Phakopsora pachyrhizi* is a biotroph, or better described as one of the fungal pathogens which requires living host cells for survival or reproduction.

Fungal pathogens that cause rust diseases can be autoecious where all the stages of the fungus occur on the same host, or heteroecious where two host plant species are required to complete the lifecycle (Alexopoulos, *et al.* 1996). *P. pachyrhizi* has a wide host range (see below) that could sustain the telial or uredinial stage. However, no hosts have been reported for either the aecial or the pycnial stage, suggesting that it may be autoecious as its uredinia and telial stages have been reported on a single host (Green, 1984).

Phakopsora pachyrhizi reaches optimum growth potential at 15-28 °C and a high level of relative humidity (free moisture) for a period of 6-12 hours (Melching, 1983). These conditions are the most conducive for disease development. Urediniospores are the most common spore type found during the growing season. These urediniospores are the primary inoculum source and with prevailing winds and conducive environmental conditions can initiate soybean rust epidemic (Yeh *et al.*, 1982b; Bromfield, 1984). The telial stage on the other hand has been induced under laboratory conditions and has on occasion been seen in the field, usually towards the end of the growing season (Yeh *et al.*, 1981a; Yeh *et al.*, 1981b; Yeh *et al.*, 1982a; Yeh *et al.*, 1982b; Bromfield, 1984).

Teliospores are generally over-seasoning structures, and have been germinated under laboratory conditions to produce basidiospores (Saksirirat and Hoppe, 1991); however, the importance of the telial stage in the development of soybean rust in the field is unknown. Teliospores are not generally considered the primary source of inoculum and are not often observed in the field (Bromfield, 1984; Ono *et al.*, 1992).

No over-seasoning stage is currently known for soybean rust, and survival of the rust requires living plant host tissue. Without knowledge of which host plants (if any) are attacked by the basidiospores or teliospores, much about the biology of this soybean rust pathogen will remain unknown, including whether the rust is autoecious (single host – rust cycle) or heteroecious (two host – rust cycle) (Bromfield, 1984).

Disease Cycle

Primary inocula for new infections are urediniospores. Epidemics of Asian SBR are characterized as having multi-cycle foci (Bromfield, 1984). After initial infection with urediniospores and through direct penetration of leaf epidermal cells, new uredinia develop in 5-8 days with temperatures between 15-28 °C along with 6-12 hours of free moisture (Marchetti *et al.*, 1975; Marchetti *et al.*, 1976; Koch *et al.*, 1983). Urediniospore production begins in as little as 9 days after infection, and uredinia can produce spores for 3-4 weeks under optimal conditions of temperature and humidity. Marginal uredinia may continue forming for an additional 8 weeks following initial innoculation (Marchetti *et al.*, 1975; Marchetti *et al.*, 1976; Koch *et al.*, 1983). Uredinia are found on both the upper and lower leaf surface, but are more common on the lower leaf surface.

This cycle repeats on the same, nearby and distant plants with new infestations as long as the environmental conditions are conducive. Wind dissemination of urediniospores facilitates short- and long-distance spread of the pathogen (Marchetti *et al.*, 1975; Marchetti *et al.*, 1976; Koch *et al.*, 1983). New disease foci can develop as long as living host plants are available. Urediniospores infect native hosts (legumes, volunteer host plants and weeds) allowing the disease cycle to continue. The disease may occur year-round in tropical and subtropical climates (Yang *et al.*, 1990).

For most rusts, teliospores are over-seasoning structures but their role in the epidemiology of Asian SBR is not well documented (Bromfield, 1984). Once present in the United States, Soybean rust will likely persist on leguminous host plants in the southern tier of States (Yang *et al.*, 1990).

Hosts

Phakopsora pachyrhizi infects 31 species in 17 genera of legumes, and 60 species in 26 other genera have been infected under controlled conditions (Sinclair and Hartman, 1995). This unusually large host range may be due to the unique ability of the fungus to directly penetrate host epidermal cells through the formation of an appressorium and germtube (Marchetti *et al.*, 1976; Keogh *et al.*, 1980; McLean and Byth, 1981; Koch *et al.*, 1983; Koch and Hoppe, 1987; Koch and Hoppe, 1988).

> **Natural hosts** (those hosts from which *P. pachyrhizi* has been found *in situ*) include species in the Fabaceae family, sub-family Papilionoideae: *Crotalaria* spp., *Desmodium* spp., *Glycine* spp., *Kennedia* spp., *Lablab purpureus*, *Lupinus* spp., *Macroptilium* spp., *Melilotus officinalis*, *Neonotonia wightii*, *Pachyrhizus erosus*, *Phaseolus* spp., *Pueraria lobata*, *Sesbania exaltata*, *Trifolium incarnatum, Vicia dasycarpa, Vigna* spp. (Bromfield, 1984).
>
> **Potential hosts** (those shown through artificial inoculations under greenhouse or natural conditions to infect and reproduce) include: *Alysicarpus glumaceus*, *Cajanus* sp., *Cajanus cajan, Calopogonium muconoides*, *Canavalia gladiata, Canavalia maritima, Centrosema pubescens, Crotalaria anagyroides, Crotalaria dissaromoensis, Crotalaria linifolia, Crotalaria pallida, Delonix regina, Desmodium discolor, Desmodium rhytidophyllum, Desmodium triflorum, Desmodium varians, Dolichos axillaris*, *Glycine canescens*, *Glycine clandestina*, *Glycine falcata, Glycine latrobeana, Glycine soja, Glycine tabacina, Glycine tomentella, Hardenbergi violecea, Kennedia coccinea, Kennedia prostrata, Kennedia rubicunda, Lespedeza bicolor, Lespedeza juncea, Lotus americana, Lotus major, Lotus purshianus*, *Lupinus angustifolius, Lupinus hirsutus*, *Macroptilium atropurpureum, Macroptilium bracteatum, Macroptilium lathyroides, Macrotyloma axillare*, *Medicago arborea*, *Melilotus officinalis*, *Melilotus speciosus*, *Mucuna cochinchinensis*, *Phaseolus coccineus, Phaseolus lunatus, Phaseolus vulgaris*, *Pisum sativum*, *Psoralea tenax, Pueraria lobata*, *Rhynchosia minima*, *Sesbania exaltata*, *Sesbania vesicaria, Teramnus uncinatus, Trifolium incarnatum*, *Trigonella foenum-graecum*, *Vicia dasycarpa*, *Vigna mungo, Vigna radiata* and *Vigna unguiculata* (Bromfield, 1984); *Alysicarpus vaginalis*, *Cassia occidentalis*, *Clitoria ternatea*, *Coronilla varia*, *Crotolaria spectabilis*, *Kummerowia stipulacea*, *Kummerowia striata*, *Lupinus albus*, *Lupinus luteus*, *Sesbania sericea*, and *Trifolium repens* (Rytter *et al.*, 1984); *Psophocarpus tetragonolobus*, *Vicia faba*, *Vigna luteola*, (Poolpol and Pupipat, 1985); *Glycine argyrea*, *Glycine curvata*, *Glycine cyrtoloba*, *Glycine latifolia* and *Glycine microphylla* (Hartman *et al.*, 1992).

The preceding species have been identified as potential hosts in greenhouse inoculations. Empirical evidence suggests that several species in the list of potential hosts are also natural field hosts (Bromfield, 1984; Hartman *et al.*, 1992; Poolpol and Pupipat, 1985; Rytter *et al.*, 1984). Additional research is needed to determine the susceptibility of other legumes native to the United States.

Identification

The identification of *P. pachyrhizi* is complicated by the morphological similarities between *P. pachyrhizi* and *P. meibomiae*. When comparing fungal morphologies, the telial stage is needed for definitive identification to species (Ono *et al.*, 1992), and this stage is not often observed in the field (Yeh *et al.*, 1981a; Yeh *et al.*, 1981b; Yeh *et al.*, 1982; Poolpol and Pupipat, 1985). Additionally, early symptoms of rust may be confused with those of other diseases such as bacterial pustule (caused by *Xanthomonas axonopodis* pv. *glycines*) (Bromfield, 1984; Sinclair and Hartman, 1999; Caldwell and Laing, 2002; United Soybean Board, 2002).

Isozyme analysis (Bonde *et al.*, 1988) supported the identification of *P. pachyrhizi* and *P. meibomiae* as separate species. When multilocus banding patterns were compared among Asian and New World rust isolates, no differences were found among the Asian isolates, nor among the New World isolates. However, the two groups differed greatly from one another, with the maximum coefficient of similarity estimated at 0.07 (7% of alleles in common) for the 14 loci examined. While isozymes represent an important research tool, they are generally not considered appropriate for use as a diagnostic test.

Classical and real-time PCR protocols have been developed to discriminate between the two species (Frederick *et al.*, 2000; Frederick and Snyder, 2001; Frederick *et al.*, 2002), and these protocols are currently being validated by CPHST's National Plant Germplasm and Biotechnology Laboratory. Sequencing of the internal transcribed spacer regions (ITS) 1 and 2 revealed greater than 99% nucleotide sequence similarity among isolates of either *P. pachyrhizii or P. meibomiae*, but only 80% sequence similarity between the two species (Frederick *et al.*, 2002). Utilizing these sequence differences, primer sets have been designed to discriminate between the two species.

Signs and Symptoms

Early symptoms of infection may appear similar to those of other diseases including bacterial pustule, bacterial blight, mustard spot, or spider mite injury (Bromfield, 1984; Sinclair and Hartman, 1999; Caldwell and Laing, 2002; United Soybean Board, 2002). The most common signs associated with SBR on cultivated soybeans are 2-5 mm tan to red-brown or dark-brown lesions (Bromfield, 1984). These lesions consist of uredinia coalesced into one large lesion.

The uredinia are often found on stems, petioles and pods, but are most abundant on leaves (Bromfield, 1984). The uredinia are 100-200 μm in diameter, ostiolate (have a circular opening through which urediniospores are released), subepidermal, erumpent and are more abundant on the abaxial (lower) leaf surface (Bromfield, 1984). Under the microscope urediniospores are 10-15 μm long and 8-12 μm wide, globose to subglobose, echinulate (spiny) and light brownish-yellow (Bromfield, 1984; United Soybean Board, 2002; USDA, 2002; USDA, 2003).

The telia are crustlike and form subepidermally in and among the uredinia and at the lesion periphery (Bromfield, 1984; United Soybean Board, 2002; USDA, 2002; USDA, 2003). Teliospores are catenulate (borne in chains of 2 to 5), variable in shape (but mostly clavate, oblong or angulate), 14-30 μm long and 5-13 μm wide, yellow to brownish, turning black with age and smooth (Bromfield, 1984).

Morphological differences between *P. pachyrhizi* and *P. meibomiae* have been characterized, and the two species may be reliably distinguished by examining the teliospores (Ono et al., 1992), however telia are not normally present during the soybean growing season. The uredinia of both *P. pachyrhizi* and *P. meibomiae* characterized by peripheral paraphyses and have pale urediniospores (Ono et al., 1992). The uredinia are often found on stems, petioles and pods, but are most abundant on leaves. The uredinia are 100-200 μm in diameter, ostiolate, subepidermal and erumpent and are more abundant on the abaxial (lower) leaf surface.

As the disease pressure increases, premature defoliation occurs, the number of filled pods decreases, number of seeds per plant decreases, yield per plant decreases, the 1000-seed weight decreases, as does seed quality (Bromfield, 1984). The disease results in 10-80% yield reduction, depending on rust severity and time of initial host infection, and the environmental conditions (Bromfield, 1984).

Clinical Diagnostics

The differentiation of *P. pachyrhizi* from *P. meibomiae* is complicated by the morphological similarities between them. The telial stage is needed for definitive morphological identification to species (Ono et al. 1992), and this stage is not always observed in the field. Trained observers may identify *Phakopsora* based on uredinia, however rapid discrimination between the species requires the use of molecular techniques.

The most accurate diagnostic tests available for the identification of *P. pachyrhizi* are molecular methods based on PCR methodologies (Frederick *et al*., 2000; Frederick and Snyder, 2001; Frederick *et al.*, 2002). Real-time PCR techniques will allow for samples to be tested for the presence of *P. pachyrhizi* as spores or in infected tissue in less than a day (Frederick and Snyder, 2001; Frederick *et al.*, 2002). Examples of symptoms can be found online (http://www.aphis.usda.gov/ppq/ep/soybean_rust; also see links provided to other internet sites).

Techniques currently available for evaluating spore viability involve either bioassay (inoculating healthy plants with spores in laboratory studies) or testing spores for germination on water agar. In either case, days to weeks may be required for results.

Epidemiology

The disease is spread through airborne dispersal of *P. pachyrhizi* urediniospores. It is not seedborne- research has shown that SBR is not transmitted by infested seed or soil (Yeh, *et al.*, 1982a).

Epidemics of SBR in Africa and South America have spread rapidly following their initial detection (Akinsanmi *et al.*, 2001; Pretorius *et al.*, 2001; Rossi, 2003). Rusts that cause these types of disease epidemics are characterized by multi-cyclic foci and copious production of urediniospores (Bromfield, 1984). Once the initial infection with urediniospores is established, through direct penetration of host epidermal cells, new uredinia develop in 5-8 days. At temperatures between 15-28 °C and with 6-12 hours of free moisture, numerous urediniospores are produced and released as soon as 9 days to produce new lesions (Marchetti *et al.*, 1975; Marchetti *et al.*, 1976; Koch *et al.*, 1983). Each lesion can produce an average of more than 12,000 urediniospores in 4 to 6 weeks, with more than 400 lesions possible on heavily infested soybean leaves (Bromfield, 1984).

A single leaf with severe rust lesions, may contain an adequate mass of urediniospores to cause an SBR epidemic (Bromfield, 1984). Although theoretically possible, the infected leaf would need to be placed on live green tissue of a susceptible host, when temperature and free moisture conditions favor infection, within a period of time when the spores are still viable and able to cause infection.

Local epidemics of SBR are characterized by examining spatial and temporal aspects, including final disease ratings of research plots, the apparent infection rates, and velocity of spread. However, specific values may vary across environment, cultivar, and between growing seasons. In Australia, during three consecutive soybean growing seasons, the final mean severities in field plots inoculated at a single focus were 9.0-10.0%, 0.9-3.0%, and 0.12% during the 1975-1976, 1974-1975, and 1976-1977, respectively (Casey, 1979; as cited in Bromfield, 1984). In another single focus study, in Japan, spatial disease gradients were determined, and the final mean severities were calculated at five 5.5m intervals. The initial severities ranged from 97% near the focus to 28% at the furthest interval from the focus (Kitani and Inoue 1960; as cited in Bromfield, 1984).

Apparent infection rate (*r*) relates disease progression to time, and is often calculated from disease progress curves using van der Plank's (1963) logistic (logit-linear) model. Several authors calculate *r* for natural soybean rust populations using disease severity as a measure of disease progression, and days after planting (DAP) as a measure for time. Under field conditions in Taiwan, Tschanz and Wang (1980) found *r* values between 0.034-0.209. Yang et al. (1990) found *r* values ranging from 0.01 to 0.25 and depending on cultivar and planting date. Casey (1979; as cited in Bromfield, 1984) found rates of 0.045-0.050, 0.034-0.038, and 0.030 across years. Finally, inTaiwan, Tschanz and Tsai (1982) found rates ranging from 0.18 to 0.376 depending on cultivar and length of photoperiod. These rates are variable, and differ, at times, in degrees of magnitude. Tschanz and Tsai (1982) reported that variation across cultivars could be partly due to delays associated with physiologic development and aimed to standardize DAP through the variable relative time (RT), (RT= DAP/ days to full maturity x 100). This enabled the authors to reduce the range of *r* to 0.216-0.312 across cultivar and photoperiod.

Velocity (meters/day) relates the spatial aspects to the temporal aspects of disease progression. Velocity of SBR has been determined, based on experimental data. For example, Kitani and Inoue (1960; as cited in Bromfield, 1984) found a spread rate of 1.0 meters/day after focal initiation in Japan. In contrast, Casey (1979; as cited in Bromfield, 1984) found lower rates of 0.45, 0.20, and 0.15 meters/day, in naturally infested Australian field plots, depending on year.

Potential for Natural Movement to the United States

It is postulated that *Phakopsora pachyrhizi* could reach the United States from Africa or South America via winds from infested areas (Unpublished data, R. Magarey & S. Isard). Transatlantic and intercontinental aerial spread of some pathogens (including many rusts) has occurred in the past (Brown and Hovmøller, 2002). Coffee leaf rust reportedly crossed the Atlantic Ocean from Angola to reach Brazil in 1970. Sugar cane rust caused by *Puccinia melanocephala* was likely carried via storm winds into the Dominican Republic (Brown and Hovmøller, 2002). It is documented that on an annual basis, cereal stem rusts caused by *Puccinia* species and tobacco blue mold caused by *Peronospora tabacina*, which survive on susceptible hosts in tropical climates, move into cereal and tobacco production areas, respectively, in North America (Brown and Hovmøller, 2002). A similar situation would be anticipated for SBR as it spreads northward from equatorial areas in South America.

Figure 1 shows areas where *P. pachyrhizi* is currently found in the world. Figure 2 illustrates the global prevailing wind patterns. Based on this information, three potential scenarios may be constructed for *P. pachyrhizi* to reach the United States as a wind-borne pathogen:

Land bridge

Local wind-borne spread from area to area and country to country across the land bridge from South America, through Central America and/or the Caribbean and eventually the U.S. is highly likely. Soybeans as well as other wild and cultivated hosts of the disease are widespread throughout the region and climatic conditions are generally suitable for spread.

Prevailing winds

The probability for long-distance spread of *P. pachyrhizi* to the United States by wind will increase as soybean production areas and other host species near and above the equator become infected, increasing the spore load in the atmosphere where prevailing winds move northward.

Extreme weather events

Extreme weather events (such as a hurricane) may carry *P. pachyrhizi* from infested areas to the United States. This however, is an unlikely scenario for the

> movement from South America to the United States because out of the more than 1,260 storms that have been tracked since 1851, no storm has ever started below 7.2 degrees North Latitude (NOAA, 2003). However, extreme weather events often begin off the west coast of equatorial Africa. With the presence of *P. pachyrhizi* in Nigeria, there is a greater potential for extreme weather events beginning in equatorial Africa to carry the pathogen into the East African islands, the Caribbean Islands, or the Southeastern United States. The level of inocula available to enter the atmosphere in any location will be related to the level of infestation in the area, the conditions for epidemics, and management conditions in cultivated areas. The use of fungicides to manage the disease in cultivated areas may reduce the spore load available to enter the atmosphere.

A key aspect of understanding the potential for *P. pachyrhizi* to be introduced by wind is the viability of spores. Data are currently lacking regarding the survival of spores under conditions associated with long-distance movement in the upper atmosphere, including in particular the effects of ultraviolet radiation.

If viable *P. pachyrhizi* spores arrive in the United States, then the probability of the disease becoming established depends upon host availability, suitable environmental conditions and the pathogen's ability to sustain itself on living hosts during adverse conditions. Experience with a related pathogen, *P. meibomiae* suggests that biological and environmental requirements can be significant limiting factors for disease establishment.

Because of the presence of year-round living host tissue, *P. pachyrhizi* survives well in Taiwan, South Africa and South America (Yang *et al.*, 1990; Akinsanmi *et al.*, 2001; Pretorius *et al.*, 2001). The ability of *P. pachyrhizi* to over-season in the United States is not known. Models estimate that in the extreme southern tier of the United States, where there are potential hosts available year–round, there is an 80-100% probability of SBR establishment (Yang *et al.*, 1990). These could serve as hosts for initial infestation and become a source of inoculum for infections during subsequent soybean growing seasons (Bromfield, 1984).

P. pachyrhizi would be expected to spread north in a manner similar to that of cereal rusts caused by *Puccinia* species (Brown and Hovmøller, 2002). Each season, cereal rusts such as *Puccinia striiformis* f. sp. *tritici* migrate north from the southern to the northern provinces of China, following susceptible hosts and prevailing winds (Brown and Hovmøller, 2002). Similarly, *Puccinia graminis* moves from the southern regions of North America to the wheat belt of the central United States (Agrios, 1988). The possibility of this occurring with SBR is significant, given the number of susceptible hosts that are present throughout South, Central and North America.

Regulatory Status

P. pachyrhizi is listed as a select biological agent (7 CFR, Part 331.2. Although the organism is established in Hawaii, it is under official control to prevent its introduction into the continental U.S. and is therefore considered a quarantine pest subject to phytosanitary measures. In contrast, although *P. meibomiae* is a quarantine pest that requires action if found because its distribution in the United States is limited to Puerto Rico, yield losses due to this pathogen are considered insignificant.

Herbaceous plants of the Leguminosae family are currently prohibited entry into the U.S. except from Canada (7 CFR § 319.37-2(a) (2003)). Soybean grain, seed and meal are currently allowed entry from all countries subject to inspection for regulated pests (including noxious weeds) and prohibited contaminants.

IMPACT

Environmental Impact

Phakopsora pachyrhizi has a wide host range (see previous "Hosts" sections, including the genera *Clitoria*, *Crotalaria*, *Lespedeza*, *Lotus*, *Lupinus Sesbania*, *Trifolium*, *Vicia*, and *Vigna*, which have species listed on the threatened and endangered species list) (CABI, 2002, TESS, 2003). It is likely that as the pathogen attacks soybean production regions of the United States, new disease management programs will be implemented. This will require the development of chemical control strategies and the use of chemicals that are not currently registered for use in the US (Section 18 documents have been submitted to the EPA for emergency use of additional chemicals if necessary. See Table 3 for additional information regarding the use of chemicals).

Economic Impact (Kent L. Smith, USDA Office of Pest Management Policy)

Soybean rust is a serious disease with potentially significant consequences. For this reason, *P. pachyrhizi* is on the list of biological agents and toxins (7 CFR 331.2), in the USDA's implementation of the requirements of the Agricultural Bioterrorism Protection Act of 2002. Yield losses and increased fungicide costs could result in the loss of billions of dollars to U.S. soybean producers. The 1984 USDA-ERS studies examined many of the factors using econometric simulation models (Kuchler and Duffy, 1984; Kuchler et al., 1984). As there are no field data on the behavior of *P. pachyrhizi* in the United States, projections of its impact were extrapolated from field observations in areas where it is endemic. Based on several potential impact scenarios, these economic analyses estimated that the annual net negative impact to the U.S. economy could range from $47 million to $4.5 billion in 1984 dollars depending on disease severity and producer response to infestation. Given the basic importance of soybeans as inputs to so many industries, impacts on the U.S. economy would be expected to be widespread and overall market disruptions would be difficult to fully predict.

Updated potential yield impacts of SBR in the United States given below are based on the established yield loss projections reported by ERS in 1984 (Kuchler and Duffy, 1984; Kuchler *et al.*, 1984). The working assumptions of the 1984 reports have not changed, but the acreage, yield, and value of soybeans have been changed to reflect more recent crop production data (USDA National Agricultural Statistics Service, 2002). Yield impact in the first year after the arrival of is expected to be relatively low on a national basis, probably in the single digit range. However, individual fields, producers, and regions may suffer severe losses. It is expected that progressively greater annual impacts nationally will follow, until the *P. pachyrhizi* extends itself through the entire range of U.S. soybean production Annual yield impacts nationally, at this point, could be as high as 10%, 15%, or even higher Regional differences in yield impacts due to differing environmental conditions have been predicted (Yang *et al.*, 1991), with regions having particularly conducive environmental conditions such as the Gulf Coast of the Southeast United States, where humidity and rainfall are high and the pathogen is expected to become established, potentially suffering losses as high as 50%.

Table 1. Expected yield impact of soybean rust in the United States based on 2001 crop data

Yield impact (%)	Soybean acreage (x 1000)	Yield per acre (bushels)	Production impact (x 1000 bu)	Value per bushel ($)	Total value of yield loss (x 1000 $)
-1	74,105	39.6	-29,346	4.30	-126,187
-3	74,105	39.6	-88,037	4.30	-378,559
-7	74,105	39.6	-205,419	4.30	-883,302
-10	74,105	39.6	-293,456	4.30	-1,261,861
-15	74,105	39.6	-440,184	4.30	-1,892,279
-25	74,105	39.6	-733,639	4.30	-3,154,650

These values merely reflect the value of the potential reductions in soybean production on a national level and do not incorporate likely increased production costs associated with fungicide application. The proper application of fungicides can limit the impact of SBR. Under conditions conducive to disease development, the presence of the pathogen in the vicinity of a soybean field should be adequate incentive to begin prophylactic sprays. By the time symptoms of disease are seen, losses are likely inevitable. However, current U.S. production practices rarely involve fungicide applications for economic reasons. Last year, fungicides for protection against diseases other than SBR were applied to less than 1 percent of the soybean acreage in the Arkansas, Illinois, Indiana, Iowa, Kansas, Kentucky, Louisiana, Maryland, Michigan, Minnesota, Mississippi, Missouri, Nebraska, North Carolina, North Dakota, Ohio, South Dakota, Tennessee, Virginia, and Wisconsin (USDA, 2003). SBR-affected producers would incur either yield losses due to the disease, increased costs associated with fungicide spraying, or both. Clearly, soybean producers whose fields are in areas where the disease can sustain itself year round may be impacted more significantly than producers that contract SBR intermittently or producers who are spared the infestation.

A critical weakness of this approach to estimating economic impacts at the national level is the assumption of no price response to reduced yields from a SBR infestation. ERS studies indicate that domestic and international prices are responsive to changes in market supply. With yield losses as much as 25%, soybean producers may be able to offset production loss through higher prices. A national economic impact analysis, which does not account for a price response represents a worst case scenario for soybean producers.

We recognize that the analytical approach presented here may not be appropriate for a national impact analysis and that the former studies by ERS (Kuchler and Duffy, 1984; Kuchler *et al.*, 1984) are an example of a more comprehensive economic analysis. While the underlying theory of the 1984 analyses is still appropriate, much has changed in terms of the structure of the domestic and international soybean industry. For example, although U.S. soybean production has increased about 45 percent since the early 1980s, its dominance of global soybean production has substantially eroded. A sharp expansion in foreign soybean output, particularly in South America, has cut the U.S. share of global soybean output from a 1980-84 average of 59 percent to a 1998-2002 average of 43 percent. The U.S. share of world soybean exports has correspondingly dropped from 79 percent in 1980-84 to 52 percent in 1998-2002. Consequently, it would now take a larger change in U.S. production to get the same market price response of two decades ago. This would presumably diminish previously calculated price benefits to farmers culminating from SBR induced production losses.

There have also been changes in soybean production technologies, shifts in regional production patterns, and new commodity programs. Since the early 1980s, there has been wide acceptance of genetically modified herbicide-tolerant varieties by U.S. soybean producers. Within the United States, soybean acreage has become more concentrated in the Midwest and less prominent in the South since the early 1980s. Southern States accounted for 16 percent of U.S. soybean acreage between 1998 and 2002, compared with 36 percent between 1980 and 1984. Part of the reason is that Southern soybean yields have fallen even further behind the national average, from 7 bushels below in 1980-84 to 11 bushels below in 1998-2002. Production costs per acre are also generally higher in the South, so with lower average yields, soybean returns per bushel are already significantly less profitable in that region. This geographic shift will have a bearing on any new analysis of grower response to an SBR infestation that accounts for regional differences in yields, costs, and prices. The current soybean commodity program makes available to producers marketing loan benefits through direct loan deficiency programs (LDPs) when market prices are lower than commodity loan rates, and counter-cyclical payments (CCPs) whenever the effective price is less than the target price. For there to be any benefit to farm revenue from a higher soybean price, it would now occur only when the price is above $5.36 per bushel. At a lower price level, farmers would just exchange a higher price for a smaller LDP. Because of this safety net, farmers could be in a position where they are no better off from a price rise, but worse off because of a yield loss.

An updated, national, comprehensive economic analysis of a major U.S. infestation of SBR, requires a much more involved analysis than presented here. Similar to the 1984 research, such an analysis should account for such factors as: price elasticities, SBR treatment costs, shifts in land use among competing crops, impacts on the livestock sector, regional impacts of an SBR infestation, soybean and soybean product trade flows, and consumer effects. For example, a significant decline in U.S. soybean production would likely lead to increased soybean prices and allow many (but not necessarily all) soybean producers to not only absorb increased pest control costs (i.e., fungicides) but enjoy greater revenues than they had received prior to the infestation. However, increased soybean prices could encourage a production expansion by foreign competitors thus limiting gains of domestic soybean producers. Other sectors of the agricultural economy, such as the livestock, corn and cotton sectors, could be impacted over time by increased soybean prices. The livestock sector, which consumes a large share of soybean output, would be adversely impacted while corn and cotton acreage would be affected through the competition for land resources. If, as anticipated, SBR initially affects the southern soybean regions of the U.S., producers in these States would likely bear the greatest burden of a SBR infestation. A very comprehensive analysis would also include the potential impact of *P. pachyrhizi* on alternative hosts such as various bean crops. Under a scenario with very severe yield losses, crop insurance could provide a “safety net” for affected soybean and specialty crop producers but place additional financial burdens on crop insurance programs. Finally, the impact on consumers would likely be dependent on the availability of substitutes for soybean-based products, including imported goods. A critical part of the trade analysis involves clarifying the impact of SBR on U.S. competitors.

HANDLING OF SOYBEANS FOR EXPORT TO THE UNITED STATES

Soybean Meal

Based on firsthand observations of soybean processing plants in Brazil, (Carl Castleton, Trip Report, Jatai, Goias June 30-July 2, 2002), most of the 90-120,000 metric tons of soybean meal exported to the United States from Brazil is processed at a single facility in Jatai; the remainder is processed in a facility in Orlandia. All facilities with soybean storage have equipment to clean and dry soybeans prior to storage. The cleaning process generates foreign material comprised of leaf litter, broken grains, pod fragments and soybean hulls. Early in the harvest season more leaf litter and pod fragments comprise the foreign material. Later in the harvest season, this material will be mostly broken grain and hulls. The foreign material is stored in a separate warehouse for a minimum of 3 months and ages up to an additional 6 months before it is used as an additive or when the next harvest arrives.

The drying process entails passing soybeans through 3-4 vertical chambers that are heated by firewood. Temperatures are consistently higher than 90° C. Soybeans are held 30 minutes in each dryer, for a total of 180 minutes. The soybeans are then crushed, flaked and expanded. Soybean oil is extracted with hexane. Afterwards, the material is heated (placed in a de-solventizer and toaster at 100-110° C for 30 minutes) to remove the solvent. Next, the meal is transferred to a dryer and exposed to 110-120 C for 15-20 minutes. During the last step, the soybean meal and foreign material (see explanation below) is combined, and placed in a dryer/cooler at 110-120 C for 15-20 minutes. The foreign material is dried in a rotary dryer/grinder that is fueled by firewood. Temperatures equal or exceed 100° C. The dryer/grinder has 3 chambers and the material spends 15-20 minutes in the dryer. The dried and ground product is then mixed in the proper proportion to prepare the finished grade of soybean meal.

The process described above results in heat destruction of the propagules. The chemical and mechanical (grinding) processes also destroy spores. Thus, assuming no untreated or fresh foreign material is added afterward, soybean meal is essentially a processed product. The main source of uncertainty is our lack of knowledge regarding whether 100% of all soybean meal is always produced as described above, such that it does not include fresh foreign matter.

The quantity of soybean meal imported to the United States in the past shows an average of 63 containers per year (1140 metric tons, Table 2) imported between 1999 and 2003. This number is expected to increase substantially.

Table 2. Imports of soybean for seed, flour and meal, and other from countries with known infestations of *P. pachyrhizi* (Source: USDA-FAS-The Oilseeds Group, US Trade statistics)

	Cumulative from March of previous year to April the following year (listed)					
Grain	Average	1999	2000	2001	2002	2003
Metric tons	14008	15268	17454	9326	16264	11727
Containers	772	841	962	514	896	646
Flour & Meal						
Metric tons	1140	62	1204	449	95	3890
Containers	63	3	66	25	5	214
Other						
Metric tons	4173	2885	1634	2633	2873	10840
Containers	230	159	90	145	158	597

If *P. pachyrhizi* were to be present in soybean meal shipments despite the treatments, detection at the port of entry would difficult because of the lack of detection systems for this purpose at the ports of entry. However, it is unlikely that the meal would be moved to a suitable habitat and come in contact with host material because the soybean meal will be used as animal feed and for additional processing once it reaches the United States. It is not likely to encounter living host plant material. The likelihood of an introduction of *P. pachyrhizi,* on imported soybean meal from countries with known infestations of SBR, to cause SBR in the US is thus negligible. This is mainly due to the effects of processing (heat treatment, extraction, grinding/crushing).

Soybean Grain

Consistent with normal commercial practices, soybeans are likely to be harvested from infested areas that are managed (probably with fungicide treatments) to reduce the incidence of the disease in the field and ensure a harvestable crop. The grain intended for export to the United States from countries with SBR will be cleaned of foreign material (either in the field or in storage facilities) to meet U.S. grade requirements (Table 3). Once cleaned, the soybean grain is dry or will be dried to industry-standard moisture content and maintained at optimum humidity level (12 – 15 % RH) for at least 60 days before shipment to the United States. Untreated foreign material should not be added to the soybean grain prior to shipment to the United States (FAO, 2003).

Table 3. Grade and Grade Requirements for Soybeans (7 CFR 810.1604, Federal Seed Act)

Grades and Grade Requirements				
	Grades U.S. Nos.			
Grading factors	1	2	3	4--s4
	Minimum pound limits of:			
Minimum test weight per bushel	56	54	52	49
Damaged kernels:	Maximum percent limits of:			
Heat (part of total)	0.2	0.5	1	3
Total	2	3	5	8
Foreign material	1	2	3	5
Splits	10	20	30	40
Soybeans of other colors[1]	1	2	5	10
Other material:	Maximum count limits of:			
Animal filth	9	9	9	9
Castor beans	1	1	1	1
Crotalaria seeds	2	2	2	2
Glass	0	0	0	0
Stones[2]	3	3	3	3
Unknown foreign substance	3	3	3	3
Total [3]	10	10	10	10

[1] Disregard for Mixed soybeans.
[2] In addition to the maximum count limit, stones must exceed 0.1 percent of the sample weight.
[3] Includes any combination of animal filth, castor beans, crotalaria seeds, glass, stones, and unknown foreign substances. The weight of stones is not applicable for total other material.

If all the "Grain" and "Other" category of soybeans indicated in Table 2 were grain, then the equivalent of more than 100 containers of soybean grain have entered the US on a yearly basis (4173 metric tons). Between 1999 and 2003, the United States imported more than 14,000 metric tons of soybean grain from SBR infested countries (Table 2, USDA, Foreign Agricultural Service, US Trade Statistics, and The Oilseeds Group). Assuming that between 0.08343% and 1% of imported seed was foreign material, between 11.69 and 140 metric tons of foreign material was imported with soybean grain. This quantity is significant, but modest in terms of grain trade generally. If grain imported into the United States contains 2% treated foreign material (US Grade 2, Table

3) then as much as 83.46 metric tons of foreign material was allowed in with grain on an annual basis.

If the grain and foreign material undergoes usual post-harvest treatments consistent with normal commercial practices (which include cleaning of grain, drying, and storage) then contamination of soy bean grain with SBR pathogen is unlikely.

USDA is currently engaged in activities to refine our understanding of *P. pachyrhizi* in foreign material. PPQ officials have visited US and Brazilian facilities to obtain information on soybean grain handling activities including the receiving, cleaning, drying and storage of soy beans at the first point of entry into the grain handling systems, and the transport systems to grain export facilities and within the facilities. The reports from these visits are in draft.

Soybean seed

Seed import volumes will not be influenced by market demands as much as grain and therefore the quantities of imported seed are not expected to be as substantial as grain or increase as rapidly. Nevertheless, the imported quantity is significant as large amounts of soybean seed are imported each year (Table 2) and sources may vary depending on both market and technical factors.

All seed imported from countries infested with *P. pachyrhizi* are US Grade 1 or higher quality. The level of foreign material in seed is expected to be generally less than for grain as seed is higher value. Seed is also handled more carefully as regards temperature, moisture, and other storage/shipping conditions to optimize germination. Further, seed is less likely than grain to be shipped bulk (a greater proportion will be packaged). Seed is also treated with fungicide as a normal commercial practice prior to planting.

Research has shown that SBR is not transmitted by infested seed or soil (Yeh, *et al.*, 1982a). The disease is strictly airborne and only through the release of airborne urediniospores, from infested foreign material into an area with actively growing host plants under the proper conditions of temperature and moisture, will it be possible to initiate infection.

For seed to serve as a viable pathway for entry into the US, relatively fresh urediniopores (as “dust”) would need to be aerially dispersed from locations where seed are being stored or handled to soybeans or other hosts in the vicinity. In the case of grain, it is anticipated that large quantities of beans would be handled in open-air conditions for long periods (e.g., unloading a bulk freighter at a port). In the case of seed, it is anticipated that quantities will be smaller and conditions more controlled (e.g., unloading sacks from a container).

Summary

The establishment and rapid spread of the disease in the Western Hemisphere raises the specter of increased risk for introduction to the United States. Two main pathways of concern are: (1) natural spread, and (2) spread via trade in grain, seed, and meal. The movement of grain and seed from infested areas to non-infested areas in Europe and North America has occurred for the past several years with no record of introductions. This experience suggests that trade in grain, seed, and meal may not provide a significant pathway for the introduction of the disease and that perhaps normal industry practice provides a high level of risk mitigation.

There is no question that the introduction of the disease into the US will impact US soybean production. Direct and indirect economic impacts are expected to be substantial. The extent of environmental impacts is less certain but a very large increase in the amount of fungicide used in the U.S. is anticipated at least for the near term until resistant varieties can be developed and marketed.

P.pachyrhizi
P.meibomiae

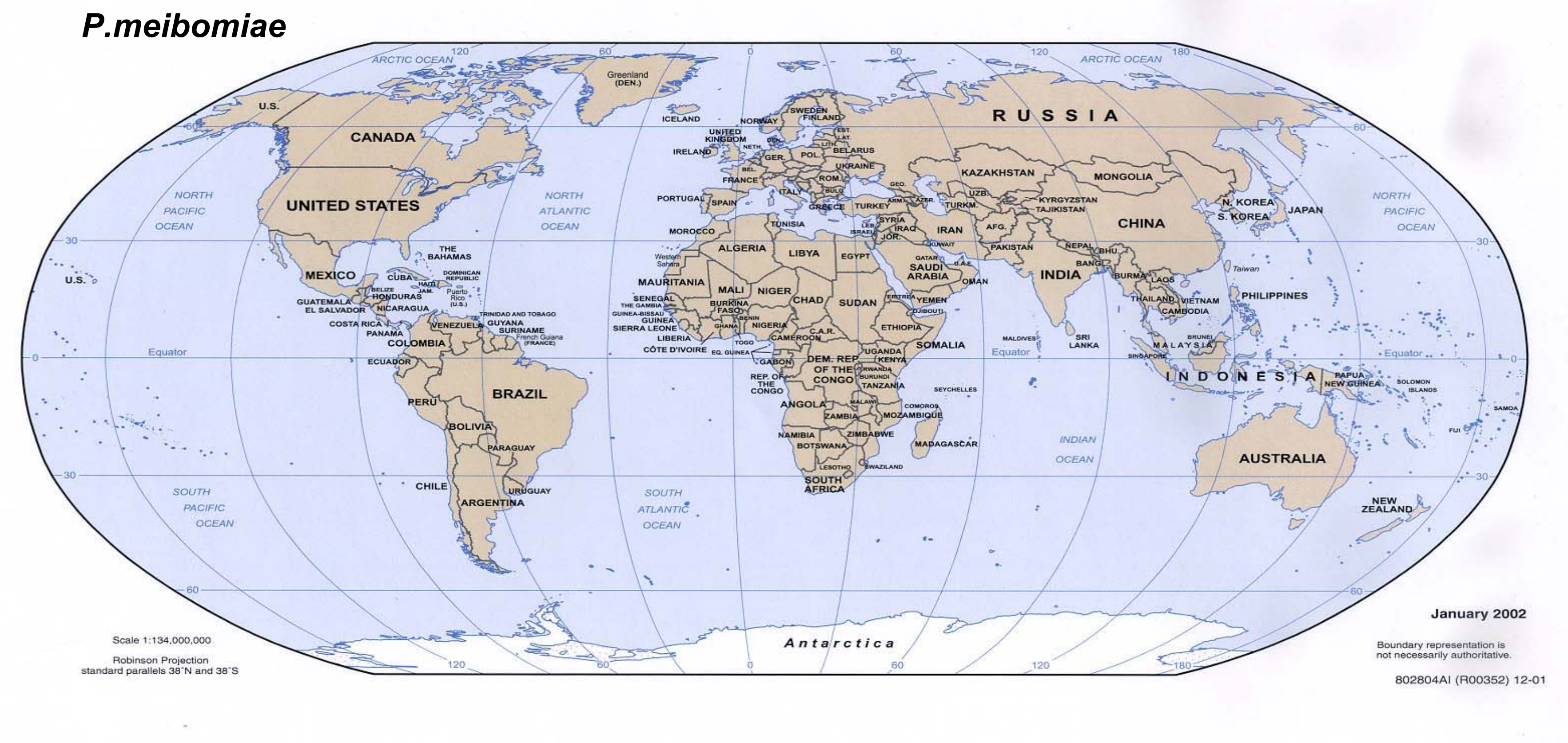

Figure 1. World distribution of soybean rust (Frederick, MD, Soybean Rust Diagnostic Workshop presentation 30 April 200).

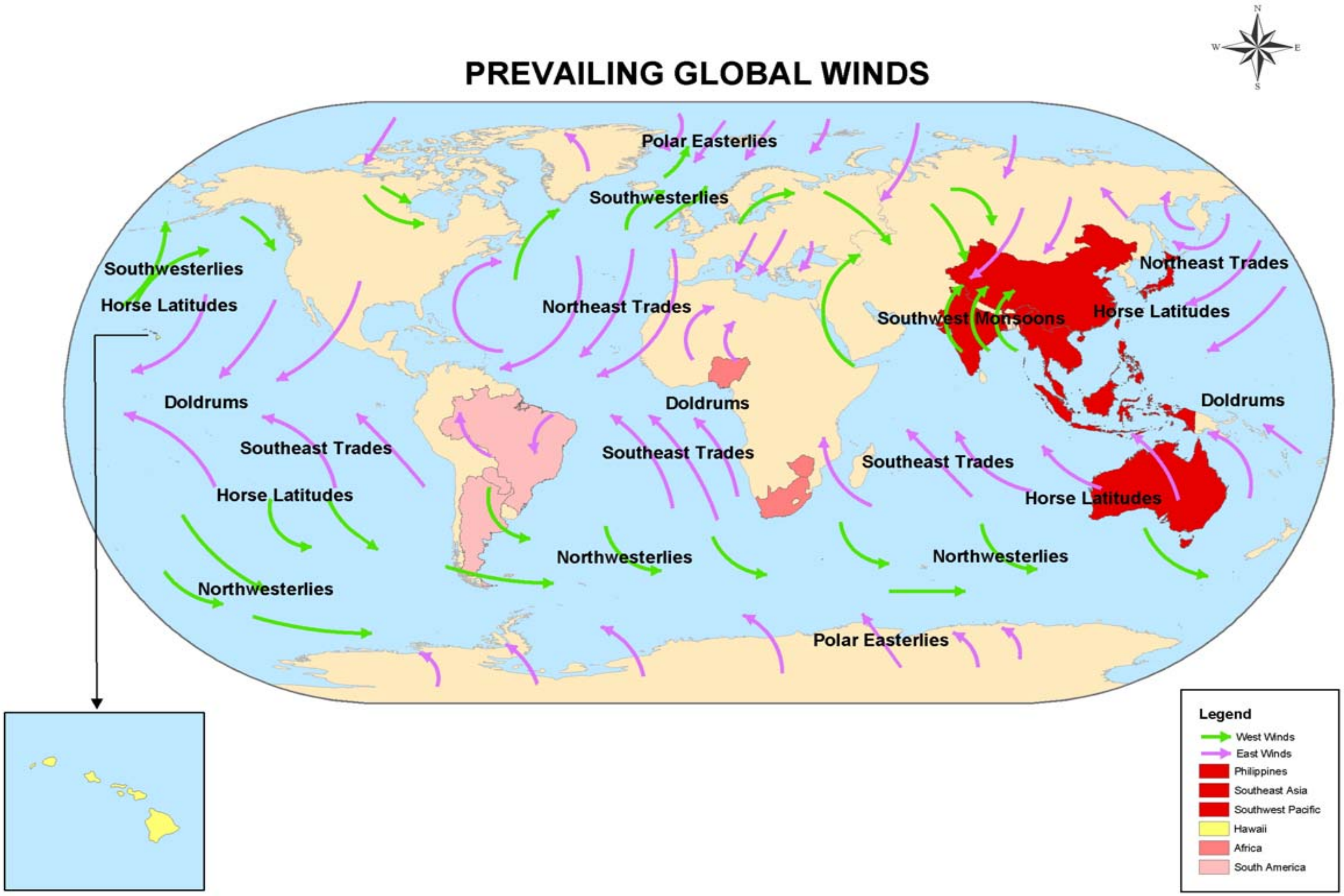

Figure 2. Global prevailing wind patterns, with potential natural pathways for *P. pachyrhizi* introduction.

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