

North American Protocol for Soybean Rust and Soybean Aphid Monitoring in Sentinel Plots

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Sentinel plots have been funded by the US Department of Agriculture (USDA), the United Soybean Board (USB), and the North Central Soybean Research Program (NCSRP) for 2006. The USDA program involves 35 states and the USB/NCSRP program includes 15 states (Table 1); a total of 35 states will have sentinel plots for monitoring soybean rust (SBR) in 2006. Five Canadian provinces are also involved in the monitoring effort this year. Some states will have a single leader for the sentinel plot program while in other states the responsibility may be shared among multiple individuals. A single SBR monitoring protocol has been developed for the USB/NCSRP, USDA, and Canadian plots. Data from all sentinel plots will be uploaded to the USDA - Legume Pest Information Platform for Extension and Education (PIPE) website.

There are three important functions of the sentinel program for monitoring soybean rust. The primary function is to serve as a warning network for tracking the spread of the disease in North American soybean production regions. For this reason and because the pathogen can only over-winter in subtropical regions, southern and Mississippi Valley states have higher numbers of sentinel plots relative to their soybean acreages than states in other regions. The second function is to quantify the timing and amount of spore production in over-wintering and growing season source areas, an important input for the soybean rust aerobiology prediction system. A third function of the sentinel plot system is to collect data for epidemiological research. For this reason, sentinel plots should be maintained after first detection unless other considerations dictate otherwise. States are encouraged to establish sentinel plots above the USDA and USB/NCSRP allotments (Table 1). Non-soybean hosts including other legumes (e.g., common bean, lima bean, lentil, chickpea) and kudzu may also be planted in sentinel plots.

In addition to monitoring for soybean rust, the Legume PIPE project will also pilot incorporation of monitoring for another pest (soybean aphid) and a new cropping system (dry beans) during 2006.

1. Number of sentinel plots

1.1. Table 1 provides the number of USDA and USB/NCSRP funded sentinel plots for each state.

2. Cultivar/Variety selection in sentinel plots

2.1. Plant soybean or common bean varieties/cultivars adapted for each geographic location.

2.2. Plant at least 2 maturity groups, early and later maturing soybean cultivars, in locations south of US Interstate 90. Sentinel plots located north of US I90 or west of US I35 may be planted to a single maturity group. However, planting soybean from more maturity groups than the minimum is encouraged.

2.3. The above variety guidelines only apply to sentinel plots that are monitoring for soybean rust. Some states will be including soybean aphid only sentinel plots and the variety and plant maturity needs may differ from soybean rust requirements.

3. Sentinel plot size and planting date

- 3.1. Sentinel plots should be at least 25' x 50' for each cultivar/host. Larger plots are encouraged, especially if deer are likely to cause damage to the crop.
- 3.2. Multiple planting dates are encouraged, but not required. Sentinel plots should be planted as early as feasible for the specific geographic region.

4. Frost and insect control measures in northern states

- 4.1. Sites should be protected against frost with row cover sheets or with other means where it is warranted. A second planting is recommended where sites are seriously damaged by frost.
- 4.2. Treat plots with insecticide where bean leaf beetle reach established treatment threshold levels.
- 4.3. Consider treatment with Cruiser Maxx on seed for seedling disease and insect control in early planted soybean plots. However, this treatment option may not be appropriate for those situations where a sentinel site is also scouted for soybean aphid, as it will remove early season aphids for up to 40 days after planting.
- 4.4. States that are also using plots for soybean aphid monitoring should communicate with their extension entomologist counterpart prior to insecticidal applications. Some insecticidal treatments may also adversely effect aphid populations and this factor should be considered.

5. Insect and nematode control measures in southern states

- 5.1. An in-furrow, at-plant seed treatment is recommended for thrips and nematode control.

6. Scouting interval, Soybean Rust (SBR)

- 6.1. Scout at a 2 week staggered interval until: 1) first bloom, 2) SBR is reported in region, or 3) environmental conditions become conducive to rust development. Scout weekly after one of the above conditions is met. After plots reach R6, scouting intensity may be scaled back.

7. Scouting interval, Soybean Aphid (SBA)

- 7.1. Scout as above for soybean rust until 1) SBA is reported in the region, or 2) soybean plants are in the late vegetative (pre-reproductive) to early reproductive (i.e. flowering) growth stage. Soybean aphid may reach threshold in some instances prior to the reproductive growth stage, but current research data suggests that treatment during this stage is not likely to result in an economic return. In most cases, weekly scouting will be necessary for no more than 8-10 weeks. Individual state entomology extension specialists will determine when weekly scouting is warranted.

8. Pre-detection SBR monitoring protocol

- 8.1. Scout for the presence of SBR by arbitrarily collecting or observing a minimum of 100 leaflets from the lower canopy (oldest, main-stem terminal leaflets) at each site. If a site has multiple cultivars, collect leaflets from the earlier cultivars

until the later cultivars reaches reproductive stage. Thereafter collect at least 100 leaflets including some from all the cultivars.

- 8.2. Incubation of legume leaves for 24-48 hr in a plastic bag containing a moist paper towel and/or inspection of leaves under a dissecting microscope (40-60X) is recommended for early detection of SBR.
- 8.3. All data from sentinel plots must be entered into the USDA PIPE national database. When uploading data, be sure to include date, plot id, cultivar, number of leaflets examined, plant stage, and disease assessment (presence, incidence, and/or severity).

9. Confirmation of SBR positives

- 9.1 The first suspected positive find of SBR in a state must be sent to the USDA lab (Dr. Mary Palm, Beltsville, Maryland) for confirmation for each host (see Federal/State Responsibility for Identification of *Phakopsora pachyrhizi*, USDA-APHIS-PPQ, December 6, 2004, reissued February 10, 2005. (Appendix B or http://www.aphis.usda.gov/ppq/ep/soybean_rust/2-10policy.pdf).
 - 9.1.1. The first positive find in every county in each state will be confirmed by a diagnostic specialist associated with the recognized NPDN lab. All confirmation will be done with approved appropriate diagnostic methods in accordance with the diagnostic SOP (see attached).
 - 9.1.2. The first suspected positive find of SBR by visual inspection in each sentinel plot should be confirmed by diagnostic clinic or state designated expert.

10. Pre-detection of SBA monitoring protocol

- 10.1. When scouting for SBA, select 20 plants at random, each from a different location (not consecutive down the row) so that the 20 plant-sample is representative of the entire variety/cultivar. Identify the average growth stage.
- 10.2. Examine the entire plant beginning with the growing point (newest trifoliolate) for soybean aphids. If plants are in vegetative growth (no pods or flowers) generally only the growing point needs to be examined. As flowering and pod set occur, examine the entire plant, including pods. Spend no more than 30 sec to examine an individual plant.
- 10.3. Unusual aphids should be submitted to your local National Plant Diagnostic Network (NPDN) lab for specimen preparation and initial screening. Please see the NPDN Soybean Standard Operating Procedure (SOP) for more information. Dr. David Voegtlin, Illinois Natural History Survey, will serve as an expert taxonomic confirmation specialist for soybean aphid or unusual aphids detected through this program. If any unusual or suspect exotic aphids are detected, Dr. Voegtlin will appropriately communicate with 1) the NPDN and 2) Dr. Gary Miller at the USDA Systematic Entomology Laboratory in Beltsville, MD in order to ensure that proper procedures of exotic pest notification are followed.
- 10.4. All data from sentinel plots monitoring for SBA must be entered into the USDA PIPE national database.

11. Post-detection SBR monitoring protocols

Once SBR has been identified in a sentinel plot or commercial field, it is important to estimate the severity of the disease at larger spatial scales to enable prediction of its spread. The following post-detection SBR assessment procedures differ depending on the size of the unit being scouted.

- 11.1. Sentinel plots – Scout on a weekly basis, inspecting 10 leaflets from each canopy level (lower, mid and upper). Determine disease severity using the 0-100% severity scale (<http://aphis.zedxinc.com/sbr/SoybeanRust.pdf>) and incidence (# leaflets infected) for each canopy level. Growth stage and canopy closure (% of soil covered by the soybean canopy) are to be collected as well. More observations may be collected if desired. Scout all varieties/cultivars planted in a sentinel plot, focusing your efforts on the varieties/cultivars that becomes infected first. Maintain that focus until defoliation. If your plan is to destroy a sentinel plot once soybean rust is detected, please consider collecting disease severity data for 2 weeks post-detection. This will allow for the determination of the extent of infection event at that site.
- 11.2. Commercial fields – From a vantage point along the field border, “guesstimate” where infection in the field is likely to be most severe. Determine disease severity at this location for the lower, middle, and upper canopy (<http://aphis.zedxinc.com/sbr/SoybeanRust.pdf>). From this location, or a better vantage point, divide the field visually into 4 quadrants of approximately equal area. Estimate a representative (or average) level of disease severity for each of the 4 quadrants (none (0), low (1), moderate (2), heavy (3)). Do not consider the lower, middle, and upper canopy levels separately, make an overall judgment for each quadrant. In addition to the disease observations, note the date and physical location of the field (GPS latitude and longitude) or distance (in miles) from the nearest cross-roads. Although the disease observations for the quadrants are only very rough “guesstimates”, when they are coupled with the observations from the heavily infected site in the same field and the field location data, they become extremely valuable for calibrating the aerobiology model and for estimating the aerial extent of the disease during the growing season.
- 11.3. County level assessment – Estimate the percentage of soybean or other legume acreage infected in county (based on specialist’s perception). Also estimate the percentage of the total soybean or other legume acreage, not the infected acreage, treated with a fungicide (based on specialist’s perception). Please note that these county-level disease observations are important for calibrating the aerobiology model and estimating the aerial extent of the disease during the growing season.
- 11.4. All data from post-detection surveys must be entered into the USDA PIPE national database.

12. Post-detection of SBA monitoring protocol

- 12.1. Scout weekly either 1) once SBA has been detected in your region or 2) at the determination of your local state entomology extension specialist. As management options during only the reproductive (i.e. flowering) growth stage appear to be economically beneficial, weekly scouting primarily at this time may only be warranted.
- 12.2. Count aphids per plant when they are below 250. Apterous (wingless) aphids are assumed to be present. Note whether or not alate (winged) aphids were also observed. If only winged aphids are present (very rare, but possible), indicate this information in the space for optional notes. Notes could also be used to mention if any predators or parasitized aphids (mummies) are present or other general relevant observations.
- 12.3. Even though an actual average number should be provided by observers for counts below 250, within-field aphid numbers will be mapped in the following

categories 1) 0 2) 1-39 3) 40-149 4) 150-249 5) 250-499 and 6) 500 and above. It is recommended that 20 plants are randomly scouted due to the potential for within-field variability for aphid populations. If an average number of aphids per plant is collected for less than 20 plants, please indicate the number of hosts scouted in the optional notes section. Categorizing aphid numbers as 250-499 or 500 and above may be done by an estimate.

12.4. All data from sentinel plots monitoring for SBA must be entered into the USDA PIPE national database.

13. Protocol for SBR monitoring in kudzu and other non-soybean hosts in sentinel plots

13.1. Scout kudzu and other non-soybean hosts such as clovers, lupine and other plants throughout the season. These hosts may be included in sentinel plots. Monitor every 1-2 weeks in sentinel plots. Indicate the presence (yes or no) and if appropriate assess disease incidence (% of plants infected) or severity (% leaf area infected). Date, location name, and detection method must be recorded. Remember that first suspected positive finds of SBR on new hosts must be sent to the USDA national diagnostic lab (Dr. Mary Palm, Beltsville, Maryland)

14. Assessment of other diseases on SBR hosts

14.1. Assessment of the following soybean disease/organism is encouraged: Septoria brown spot, frogeye leaf spot, target spot, downy mildew, Cercospora blight, bacterial pustule, and bacterial blight. Assessment of the following common bean disease/organism is encouraged: common rust, common bacterial blight, *Bean common mosaic virus* (and aphid vectors), *Curly top virus* (and leafhopper vectors). Record presence/absence of the disease/organism. Disease incidence and/or severity ratings may also be collected at the cooperator's discretion. If incidence and/or severity data is collected, the same rating scales used for soybean rust is to be used. Additional disease damage and other pertinent information may be recorded in the notes field on USDA PIPE data entry forms.

15. Assessment of other insect pests on SBA/SBR hosts

15.1. It is recommended that notes of presence and/or infestation levels of other economically important pests of soybean and other legumes such as common bean also be mentioned by observers. Bean leaf beetle is an important pest of soybean and Mexican bean beetle and leafhoppers are important pests of common bean; and information regarding these pests could be beneficial and utilized by the local state extension specialist in their commentary to growers.

16. Destruction of sentinel plots infected with SBR

16.1. The decision to spray or destroy sentinel plots after SBR detection is the responsibility of individual states. If the decision is made to destroy a plot the eradication date must be uploaded to the USDA PIPE database. This information will give modelers a better idea of the level of SBR inoculum production in a geographic area.

16.2. If the sentinel plot has been destroyed due to SBR, please inform your extension entomology counterpart as this will also end SBA monitoring.

17. Data input to USDA-PIPE website

- 17.1. Please upload data in a timely manner consistent with scouting frequency. Data for the first find of soybean rust in a state in 2006 should be submitted within 24 hours of final confirmation. (Note: The first suspected positive find of SBR in a state must be sent to the USDA lab, Dr. Mary Palm, Beltsville, Maryland, for confirmation for each host). Data for the first find of soybean rust in a county in 2006 should be submitted within 72 hours of final confirmation. More timely data submission is always encouraged.
- 17.2. Observers will be provided with user ID and password to access the USDA PIPE observer website interface.
- 17.3. Observations can be uploaded using an on-line form, by uploading an excel file (template provided), or by PDA (as download from observer site).
- 17.4. Disease severity should be assessed using a 0-100% severity scale. However, because most leaves drop when severity reaches around 50 percent and the assessment does not take into account necrotic tissue, most values will fall within 0-50% range. Photographic definitions are available by clicking images on USDA PIPE website (<http://aphis.zedxinc.com/sbr/SoybeanRust.pdf>).
- 17.5. Multiple protocols are available for uploading data for SBR. The **pre-detection** protocol is for general use for sentinel plots prior to detection. The **post-detection** protocol is used for sentinel plot post detection and for epidemiological data.
- 17.6. Multiple protocols will also be available to upload SBA data. There will be a sentinel plot, a research or ad-hoc, and an available upload for other mobile scouting data.
- 17.7. Observers are encouraged to participate in training sessions/workshops for using the web tools. A help document is also available as an aid for filling out the observation forms.
- 17.8. All technical questions regarding this protocol and data entry should be addressed to database manager, Julie Golod (golod@zedxinc.com).

Table 1. Soybean and kudzu statistics and sentinel plot allocations by state and funding source for 2006.

State	Soybean Acres ^a	Planting Date ^b	Kudzu Acres ^c	USDA Sentinel Plots		USB/NCSRP Sentinel Plots
				Total	Over-wintering ^d	
Alabama	150	May 25 to June 25	117510	22	10	
Arkansas	3030	May 25 to June 20	10091	20		
Delaware	185	May 28 to June 26	1	5		
Florida	11	May 15 to June 15	12449	30	15	
Georgia	180	May 27 to June 27	151318	22	10	
Illinois ^e	9500	May 15 to June 9	529	10		10
Indiana ^e	5400	May 15 to June 5	98	10		10
Iowa ^e	10100	May 14 to June 2		10		10
Kansas ^e	2900	May 25 to June 20	6	5		15
Kentucky ^e	880	May 25 to June 25	18051	15		
Louisiana	480	May 15 to June 15	4824	25	10	
Maryland	495	May 28 to June 26	134	5		
Michigan ^e	2000	May 18 to June 3		5		15
Minnesota ^e	6900	May 16 to June 3		5		15
Mississippi	1600	April 25 to May 25	250632	25	10	
Missouri ^e	5000	May 25 to June 1	1166	15		5
Nebraska ^e	4700	May 18 to June 4	51	5		15
New Jersey	95	June 14 to July 14	9	5		
New York	190	No data		5		
North Carolina	1500	May 20 to June 30	57660	15		
North Dakota ^e	3000	May 19 to May 29		5		15
Ohio ^e	4500	May 10 to June 7	58	10		10
Oklahoma	320	May 18 to June 22	31	5		10
Pennsylvania	440	May 20 to June 10	1	5		5
South Carolina	430	May 27 to June 27	73742	15	2	
South Dakota ^e	3900	May 20 to June 6		8		15
Tennessee	1130	May 30 to June 25	64862	15		
Texas	260	May 3 to June 14	50817	10	5	5
Virginia ^e	530	May 20 to June 30	11357	10		
West Virginia	19		1318	5		
Wisconsin ^e	1610	May 15 to June 20		5		10
Total	71435		826717	347	62	165

^a Acres planted (x 1000), National Agricultural Statistics 2005.

^b National Agricultural Statistics, 2002

^c Acres, Daryl Jewett, APHIS.

^d Over-wintering sentinel plots are likely to be carried over into the growing season.

^e Some funds available for soybean aphid monitoring.

Table 2. Edible dry bean acres and associated USDA sentinel plots for 2006.

State	Dry Edible Bean Acres ^a	Sentinel Plots
Colorado	130	8
Idaho	100	9
Oregon	7.5	9
Washington	48	9
Total	285.5	35

^a Acres planted (x 1000), National Agricultural Statistics 2005.

Table 3. Canadian Sentinel Plots

Province	Hosts – Acres Acres	Planting Date	Sentinel Plots
Ontario	Soybeans - 2500 ^a	May 10 to May 30 ^b	30
Québec ^d	Soybeans - 450 ^c		35
Alberta	Common Bean		1
Manitoba	Common Bean		1
Saskatchewan	Common Bean		1
Total	2950		68

^a Acres planted (x 1000). Albert Tenuta, OMAFRA.

^b Albert Tenuta, OMAFRA

^c Acres planted (x 1000). Claude Parent, DIST.

• Mobile teams must seek diagnostic confirmation of suspected positive observations if soybean rust has not had previous laboratory confirmation in that state. New state confirmations should be confirmed by sending samples to the APHIS-PPQ National Identification Services, but only through the State and/or Land Grant University (NPDN) lab as a first screen (see Federal/State Responsibility for Identification of *Phakopsora pachyrhizi*, USDA-APHIS-PPQ, December 6, 2004, reissued February 10, 2005. (Appendix B or http://www.aphis.usda.gov/ppq/ep/soybean_rust/2-10policy.pdf).

Ongoing laboratory diagnosis can be provided by state and/or Land Grant University National Plant Diagnostic Network (NPDN) labs.