SORTING THROUGH THE SOYBEAN LEAF DISEASE COMPLEX[1]

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Although soybean rust was not detected in Wisconsin n 2005, Plant Disease Diagnostics Clinic (PDDC) staff spent a substantial portion of their time this past growing season examining soybean samples for evidence of soybean rust. The basic strategy for monitoring samples for soybean rust at the PDDC uses an initial visual and microscopic examination of putative rust samples, with a follow-up where appropriate with a more sensitive technique called polymerase chain reaction (PCR).

Visual examination of rust samples is relatively straightforward. Soybean leaves [and leaves of other potential hosts of *Phakopsora pachyrhizi* (the soybean rust pathogen) such as snap bean, pea, bird's-foot trefoil, white clover, purple crownvetch, lupine, and yellow sweetclover] are initially scanned with the naked eye for brown or yellow spots. Suspect areas are then viewed under a dissecting microscope at magnifications that range from roughly 10 to 60 times normal size. In particular, one looks for rust pustules, the pimple or volcano-like reproductive structures of the soybean rust fungus that form on the undersurface of leaves. If pustules are not present, leaves are placed in a moist chamber [i.e., a plastic bag or other container (e.g., a petri-plate) lined with moistened paper toweling] for 24 hours, then reexamined. All soybean samples submitted to the PDDC in 2005 tested negative for soybean rust, showing no signs of *P. pachyrhizi* sporulation.

Confusion regarding the presence of soybean rust arises because several common soybean diseases mimic soybean rust. The most common of these diseases is brown spot, caused by the fungal pathogen *Septoria glycines*. This disease leads to the formation of numerous small brown spots on soybean leaves, particularly those from plants that are under stress. Brown spot can be distinguished from soybean rust as *S. glycines* does not produce pustules, but does produce urn-shaped reproductive structures filled with spaghetti-like spores that are readily visible under a compound microscope. Another common soybean rust look-a-like is bacterial blight. This disease is caused by the bacterium *Pseudomonas savastanoi* pv. *glycinea*, which causes small, angular leaf spots with yellow haloes. *P. savastanoi* pv. *glycinea* does not produce pustules or other reproductive structures, but under the compound microscope, one can often observe large numbers of bacterial cells streaming from bacterial blight leaf spots. Probably the most problematic soybean rust look-a-like is downy mildew, which causes yellow to brown spots on soybean leaves. To make matters even more confusing, the downy mildew pathogen (*Peronospora manshurica*) sporulates on the undersurface of leaves forming masses of spores that look very similar to the spore masses produced by *P. pachyrhizi*. Microscopically however, *P. manshurica* produces oblong "spore" (technically called

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sporangia) that are borne on antler-like structures, rather than spores borne in pimple-like pustules.

If soybean rust-like spores are observed on soybean (or other) leaves, definitive confirmation of *P. pachyrhizi* requires use of polymerase chain reaction (PCR). PCR is a techniques that allows one to look for a sequence of DNA (i.e., genetic material) that is unique to a particular organism (in this case *P. pachyrhizi*), even when this sequence may be present in very low numbers. PCR technology uses enzymes that naturally replicate DNA in cells to make copies of the unique DNA sequence of interest in a test tube, thus making the DNA more readily visible using light-sensitive dyes. In the case of soybean rust, first reports of the disease on soybean and other hosts in Wisconsin must be confirmed with PCR by USDA APHIS. All subsequent finds on a particular host need not be confirmed by PCR, but can be made using microscopic examination. However the PDDC's current policy is to confirm at least first reports (on soybean or any other host) in each Wisconsin county using PCR.

In 2006, the PDDC will continue to offer free soybean rust testing for soybeans and other soybean rusts hosts. Submission forms are available by mail from the PDDC or online at www.plantpath.wisc.edu/pddc. For additional information on soybean rust and other soybean diseases, feel free to contact the PDDC at (608) 262-2863 or bdh@plantpath.wisc.edu.