Julia Scher, Resource Coordinator, Identification Technology Program, USDA-APHIS-PPQ-CPHST

CPHST’s Identification Technology Program announces ID Source, released in September 2011 and developed through collaboration between CPHST and Colorado State University. ID Source is a searchable gateway to websites that focus on identification of plant pests, diseases and weeds, and can be used to quickly find trustworthy websites for identification as well as for screening and detection of plant pest organisms. ID Source is now available at idsource.colostate.edu.

The ID Source team evaluates hundreds of websites to determine their value for individuals associated with plant protection. Only those sites that support the identification needs of users are added and become “ID Aids.” Useful tools and information contained in ID Aid websites range from fact sheets, image galleries and keys, to map collections, molecular tools and more.

Each ID Aid is thoroughly reviewed and “described,” both to provide information to users about ID Aids as well as a way for users to search for them. ID Source provides two ways to search for ID Aids – by choosing filters, and by entering search text:

- Information about ID Aids, such as an ID Aid’s geographic region, organism focus, type of site content, and commodity focus, are assessed and recorded so that users can filter ID Aids by e.g., region, organism, or commodity.

New First Detector Site Launched

- SPN workshop at the Morton Arboretum
- National Repository update: nematode fields

Issue Highlights

- NPDN Star-D auditor training
- Diagnostic tip: isolating Septoria from tomato leaves
- Difficult/unclear data entries

Next Gen NPDN
Third National Meeting
Berkeley, California

November 6–9, 2011

New pests and pathogens emerge, techniques and protocols change: Are you Ready?
NPDBN STAR-D Laboratory Accreditation Auditor Training  

From September 26–29, 2011 nine NPDN members and three of our USDA-APHIS-PPQ-CPHST colleagues gathered at the University of Florida in Gainesville Florida for a 4-day laboratory accreditation auditor training. This training was the next step needed to continue the development of the NPDN STAR-D project.

The training was based on ISO 17025 Standards, the main standard used by testing and calibration laboratories. It covered basic internal auditing concepts and techniques and case studies giving participants the opportunity to discuss as well as apply what they learned during the training. The presentations included information on coordinating a quality management system audit, how to construct an audit program, effective auditing techniques, evaluating the significance of audit findings, methods for improving communication skills during the audit presentation, how to report the findings and conclusions, and developing and implementing corrective action programs. Our experienced instructor used real world examples, his own personal experience and hands-on exercises to train attendees to be ready to conduct impartial and effective audits.

The three days of classroom training culminated on the fourth day in a simulated audit of the University of Florida Plant Disease Clinic with laboratory manager Anne Vitoreli.

Before beginning the audit Anne told the group “I’m open to all comments and critiques to help improve the lab”. We had a chance to utilize the auditing skills we had learned over the previous three days.

Each of the three audit teams prepared a report including the team’s audit checklist, non-conformances found during the audit and additional audit findings. This information was compiled into a comprehensive report and will be presented to the Operations Committee and to Anne to help identify the strong points of the lab as well as areas that need some improvement.

Pat Shiel and Kathy Burch of USDA-APHIS-PPQ-CPHST made the arrangements for NPDN members to receive the auditor training by a certified trainer, provided by SAI Global.

The participants from NPDN came from all over the country. They included Dawn Dailey O’Brien and Karen Snover-Clift (Cornell University), Anne Vitoreli (University of Florida), Nick (Raghuwinder) Singh (Louisiana State University Agricultural Center), Ron French (Texas A & M University), Judy O’Mara (Kansas State University), Laura Jesse (Iowa State University), Jason French (New Mexico State University), Shouhua Wang (Nevada Department of Agriculture), and from CPHST; Kathy Burch, Patrick Shiel and Geoffrey Dennis. With completion of this training, attendees are now capable of conducting internal and external audits for NPDN and collaborator laboratories. This was the first of several auditor trainings planned to be offered as the STAR-D Quality Management system develops.
Have you ever tried to isolate *Septoria* from tomato leaves at the request of a colleague for a clean culture; not as easy as it sounds. The following tips may be a helpful resource to reduce contamination when isolating from foliar tissue.

1. Collect tomato leaves infected with *Septoria lycopersici*. Lyophilize for 2–3 days to fully dry the leaves. (This reduced contamination of fast growing fungi in my experience.)

2. Cut section of leaf containing *Septoria* lesion and soak in 10% bleach for 45 seconds. (I use 10 mL in a 14 mL tube.) Transfer leaf to 10 mL sterile water in a 14 mL tube and invert 5–6 times to rinse. Gently blot dry between paper towels or kimwipes.

3. In a regular petri plate, put a piece of Whatman #1 filter paper circle in the bottom and add a little sterile water, just until wet. Lay a sterile rubber band on top and then a circle of sterile screen on top of that to provide a raised platform for your leaf sample. When the lid is placed on the petri plate, this will create a humid chamber without your leaf sample sitting in water (figure 1).

4. After you’ve blotted your surface sterilized leaf sample dry, place on top of the screen, lesion side up. Put the lid on the petri plate and put all your plates in a closed bag. Incubate at 24°C overnight.

5. Under the dissecting microscope, look for spores oozing out of the lesions. Touch spores with a sterile needle then transfer spores to clarified V-8 agar plates containing kanamycin or other antibiotic (see recipe below). Incubate these plates at 24°C for several days. *Septoria lycopersici* will be dark in color with spores appearing later as small pinkish spots (figure 2).

### Clarified V-8 preparation:

1. Add 5 g CaCO₃ to 340 mL V-8 juice and stir for at least 15 minutes. Aliquot into 50 mL tubes and centrifuge balanced tubes for 15 minutes at 4,000 rpm. Carefully pour 40 mL of the clarified V-8 into 50 mL tubes for storage at -20°C. Label the tubes “5X clarified V-8” along with the volume.

2. When ready to prepare agar plates, thaw an aliquot(s) of the clarified V-8 and mix:

   - 40 mL 5X clarified V-8 juice
   - 160 mL dH₂O
   - 3 g agar
   - 0.2 g sucrose
   - 0.04 g yeast extract
   - 200 mL total volume

3. After sterilization, add 400 µL Kanamycin (25 mg/mL, filter sterilized) to 200 mL cooled media and pour plates.
Difficult / Unclear Data Entries
Nancy Gregory, University of Delaware, Chair, National Database Committee

All of us who enter data in systems that upload to the National Repository have encountered sample identifications that we do not readily know how to enter. If the software system you are using has drop-downs, you may find what you are looking for readily, or you may search around a bit before you do. You may find several ambiguous entries that you use all the time, such as “Insufficient Sample” or “Physiological Responses”. It is best to try to get the entry as accurate as possible, even though that may be frustrating at times.

To review data at later points, it will be best to have data as consistent as possible, and as specific as possible. For that reason, there are very few “Unknown” choices and it is not encouraged for you to enter “Unkowns”. There are a few “Unspecified Pathology” entries linked to fungi we encounter often, such as a Fusarium sp./spp., or Botrytis sp./spp.

There are also syndromes or common names associated with “Unidentified Agent”, for example, root rot. There are some codes for “Undetermined”, but most of those are associated with entries such as Identification Analysis for Plant ID, Weed ID and others. If you have trouble finding an entry, you may request one. In those cases, the staff at CERIS at Purdue would direct the diagnostician to an existing entry if one is already in the system. If the pest is not in the system, it can be added.

The Database Committee reviews some new requests. You can also e-mail one of the members of the Database Committee, who may have a suggestion. The Committee is in the process of updating the Upload Guidelines and that document should be available in early 2012. That document will have hints on how to enter some samples. Always ask if you have questions!

Diagnostics Committee Website Addition
Karen Snover-Clift, Department of Plant Pathology and Plant-Microbe Biology

Due to input from the members of the diagnostics program area committee, an addition has been made to the diagnostics page of the NPDN website. There was some concern about diagnosticians having questions on how to handle unknown samples that appear to be significant in nature. We have specific protocols for many of the select agents and a few significant pathogens and pests, but we do not have clear guidance on the question of unknowns. Therefore, we added a section to the top of our webpage that provides Joel Floyd’s (USDA-PPQ-PPQ, Domestic Diagnostics Coordinator) contact information.
NPDN Launches New First Detector Site!
Amanda Hodges, Department of Entomology and Nematology, University of Florida

NPDN Training and Education has a new home — go to www.firstdetector.org to check out the new site! All of the information that you previously could access on the former NPDN training site is still available on the new site. Users going to the former site (http://cbc.at.ufl.edu/) will be redirected to the new address automatically. Behind the scenes, the training database migrated to a new server. The visible changes that you will recognize on the site include more informational pages about the training program, before creating an account and logging into the NPDN training site. For example, you can read about the learning objectives of an e-learning module before logging into the site or creating an account. Once logged into the system, users can toggle back and forth between ‘workshops’ and ‘on-line training’ on the top right of your login screen in order to access the features previously available on the site after the login.

Questions or comments regarding the new site can be directed to the NPDN Training and Education Program Area Manager, Amanda Hodges achodges@ufl.edu.

SPN Workshop at the Morton Arboretum
George Hudler, Department of Plant Pathology and Plant-Microbe Biology, Cornell University

The Morton Arboretum in Lisle, Illinois was recently the site for the Midwest Sentinel Plant Network workshop sponsored jointly by the American Public Gardens Association (APGA) and the National Plant Diagnostic Network.

Dan Stern and Rachel McCarthy were the workshop organizers from their respective organizations, ably assisted by Casey Sclar and George Hudler. Stephanie Adams and Fred Miller of the Arboretum staff were most generous with their time and talent, leading two invigorating and informative “pest walks” to view the grounds and talk about pest/pathogen identification and management. Other Morton staff provided tours of the library and the
research labs, herbarium and collections. (Yes, the facilities were top notch and left most of the rest of us green with envy.) Tom Creswell (Purdue) and Nancy Taylor (Ohio State) provided valuable insights from the “front lines” of the diagnosticians on the NPDN side of the ledger, helping to ensure that as our collaboration grows, communications will leave minimal ambiguity.

Thirty four representatives from public gardens throughout the Upper Midwest (Minnesota, Wisconsin, Michigan, Illinois, Indiana, Ohio and Missouri) attended and in addition to the education which took place, productive relationships with our two previously distanced organizations were also forged. Among other things, we’re learning that a high priority in the APGA vision/mission is to play a more active role in educating botanical garden staff and the public at large about invasive pests and pathogens. Thus, they are behind the Sentinel Network initiative 100% and leave no doubt that their enthusiasm and commitment to excellence are genuine. Try to get to a workshop near you as time and resources allow, and you’ll soon find yourself as we have, caught up in their enthusiasm.

Top: participants fill out laboratory sample submission forms during the sample submission session. Bottom: Fred Miller and Stephanie Adams from the Morton Arboretum on the pest walk. Photos courtesy of George Hudler, Cornell University.

National Repository Update
Nematode Fields Now Available
Michael Hill & Eileen Luke, CERIS, Purdue University

The software for supporting three new optional nematode fields was placed into production in the National Repository. The three new fields that are available for uploading are:

1. Nematode Density Count
2. Nematode Density Unit
3. Nematode Life Stage

These fields can be uploaded using the XML upload service or by directly entering the fields using the online data entry form. The changes made to the
Data upload software are 100% backwards compatible meaning that no changes are required by the lab management systems to continue sending data in to the National Repository. For those using PDIS, these optional fields are available for entry and are part of the standard upload to the National Repository. We have provided a screen shot that demonstrates how nematode fields can be entered using the online data entry form.

Please contact Mike Hill (mhill@ceris.purdue.edu) or Eileen Luke (lukee@ceris.purdue.edu) if you have any questions regarding data uploads to the National Repository.

New ID Aids are always being added to ID Source, and the development team constantly works on improving the site’s performance by editing ID Aid data, giving users better and faster search results and enhancing search options. ID Source aims to evolve and grow for long-term success. A key element of that growth is ID Source’s users. ID Source’s vision relies on a user community that takes an active role and has a stake in its success. This initial release therefore incorporates and emphasizes ways users can participate by contributing ID Aid suggestions, rating and reviewing ID Aids, and providing feedback of all kinds. The more users participate, the more valuable ID Source will become. With users’ help, ID Source can become dynamic, self-sustaining and the first stop for online identification needs.

Users can enter text to find ID Aids by, e.g., words occurring in their title or sponsoring organization. This text search also functions as a quasi “search engine” — all ID Aids are web-crawled in order to locate scientific names and common names that appear on their web pages. For each scientific or common name found in an ID Aid website, one occurrence of it is recorded for that ID Aid — a single ID Aid may have over 1,000 such occurrences. This data then fuels ID Source’s “ID Aid Site Search,” enabling users to enter names or parts of names, and then see ID Aids in their results that contain those names.

Users may find searching for ID Aids with ID Source easy and enjoy results that are more focused and relevant than standard Internet search engines can provide.
As of October 2011, nearly 1,500 identification-themed websites have been vetted and selected as ID Aids. Thumbnail ID Aid screenshots have been captured and detailed information about ID Aids is displayed for easy evaluation while searching for the right ID Aid. We mine the Web for both domestic and international ID Aids to help address the issue of pest introductions via global trade.

Recent upgrades
- The speed of search results, within one month of ID Source’s initial release, is now twice as fast due to successful back-end performance changes.
- “Full text search” capability has been implemented to make the ID Aid Site Search more logical and user-friendly.
- ID Aid Site Search results will soon be improving as we incorporate lists of important pests into the ID Aid web crawl.

Ways to participate
- Provide your ratings and reviews of ID Aids to assist future users when they attempt to find the “best” or most valuable ID Aids.
- Suggest web sites you think would be valuable ID Aids to keep the ID Aid collection growing and current.
- Give us feedback about ID Source — it is simple to do and will help us improve and increase the value of ID Source for you and your colleagues.

Questions or comments about ID Source are welcome. Please contact: Julia Scher at Julia.L.Scher@aphis.usda.gov or (970) 490-4465.
Soilborne Plant Pathogens and California Nematology Workshop

Timothy Paulitz, USDA-ARS

Come and meet with colleagues from the western U.S. working on various aspects of soilborne fungal pathogens, nematodes and diseases — from the molecular to the applied. This meeting is very informal and loosely structured, allowing lots of time for discussions and interactions. We will have a garden tour on Tuesday March 20 at the Huntington Botanical Gardens, with Dr. Martin Stoner. The garden will be closed to the public. The California Nematology Workgroup will meet on Tuesday afternoon, March 20 and there will be a social/dinner at the garden in the evening. Research presentations will be on Wednesday, March 21 and Thursday, March 22. The focus of the symposium will be ornamental and landscape pathology, with speakers to be announced.

We are also offering two student scholarships for $500 plus free registration. The details are on the web site.

Early registration deadline is March 1, 2012. For more information, including information on registration, payment and hotel visit http://soilfungus.ars.usda.gov/. Please send the registration form to Dr. Timothy Murray, Dept. of Plant Pathology, Rm. 345 Johnson Hall, Washington State University, Pullman, WA 99164-6430 USA, or FAX to (509) 335-7674, or send scan to paulitz@wsu.edu.

Local arrangements: Ole Becker (ole.becker@ucr.edu) or Antoon Ploeg (antoon.ploeg@ucr.edu)

Program Chair: Timothy Paulitz, USDA-ARS, Pullman, WA.

Hope to see you there.

Meeting of the 58th Annual Conference on Soilborne Plant Pathogens (formerly Soil Fungus Conference) and the 44th Annual California Nematology Workshop

March 20–22, 2012

Huntington Library, Art Collections, Botanical Gardens
1151 Oxford Road San Marino, CA 91108

Upcoming Events

National Events

November 6–9, 2011
NPDN National Meeting
Berkeley, CA

November 13–16, 2011
ESA 59th Annual Meeting
Reno, NV