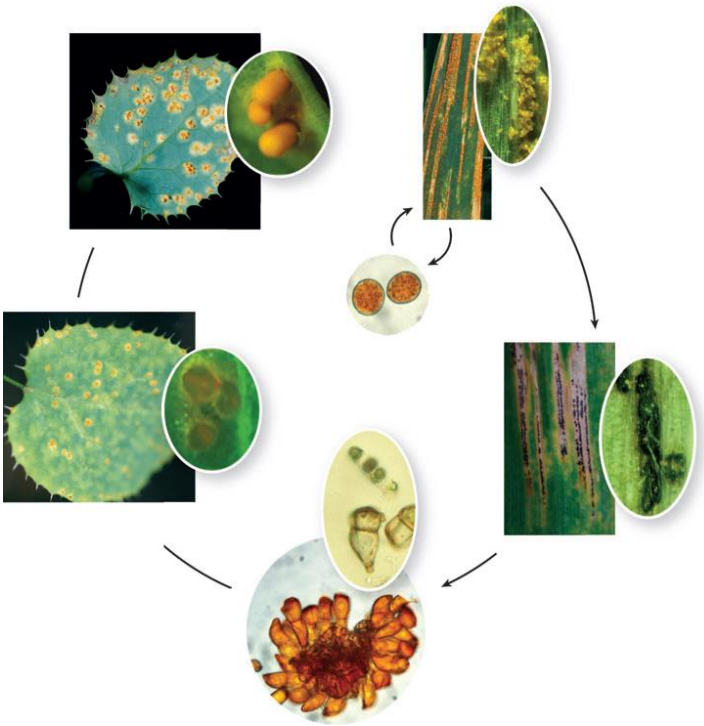


Barberry rust survey
Developing tools for data management and dissemination



Source doi: 10.1146/annurev-phyto-072910-095230

Jens G. Hansen, Poul Lassen Annemarie F. Justesen, Kumarse Nazari,

David Hodson & Mogens S. Hovmøller

11 July, 2013

Photo: Mogens Hovmøller & Jens G. Hansen (AU, Agro)

Table of Contents

Summary

The objective of this report is to document what have been carried out in the DRRW Activity 23.a.1., sub-task: "Barberry surveillance database and dissemination tools". The draft version was developed to support another DRRW project: "Berberis rust survey in the Ug99 pathway in CWANA", led by ICARDA and Aarhus University, and data from this project is used for describing the system developed. The documentation and results cover the development of a surveillance protocol, development of tools for entering and storage of molecular diagnostics data and the development of management and dissemination tools. In the report we propose improvements of the system and we discuss how to organise activities, data sharing and collaboration between partners working on barberry and other alternate hosts for *Puccinia* spp. The ultimate aim is to gain insights into the role of barberry in the epidemiology of *Puccinia* spp. infecting cereals and grasses. Therefore, in the frame of the DRRW project, we propose to use the Wheat Rust Toolbox to integrate rust on barberry survey data and pathogen monitoring data based on molecular diagnostics, infection assays and other methods from research groups world-wide in order to obtain added value of merging data and expertise.

Background

The common barberry and several other *Berberis* spp. serve as the alternate hosts to *Puccinia graminis, P. striiformis* and other *Puccinia* species. Sexual cycles on *Berberis* spp. may generate virulence combinations that could have serious consequences to cereal crop production (Yue Jin, 2011). The importance of the *Berberis* spp. as alternate host for *P. striiformis* f.sp. *tritici* in different areas is relatively unknown (Jin et al. 2010; Wellings 2011, Hovmøller et al. 2011). Previous studies of the genetic diversity of *P. striiformis* f.sp. *tritici* in Europe, North America and Australia have shown a clonal structure indicating that no sexual recombination is taking place (Hovmøller et al. 2002). However, studies of a Chinese population of *P. striiformis* f.sp. *tritici* from the Gansu province showed high genetic diversity and increased ability to produce teliospores indicating that sexual recombination may take place in this area (Mboup et al. 2009, Duan et al., 2010). Among 3,703 aecia sampled from barberry plants of three species, *B. shensiana, B. brachypoda,* and *B. soulieana*, under natural conditions in Gansu and Shaanxi provinces, four produced Pst uredinia on susceptible wheat cultivar Mingxian 169 (Zhao et al. 2013)

In the Central, West Asia and North Africa (CWANA) region, the importance of *Berberis* spp. as alternate host for *Puccinia spp.* infecting cereals is currently unknown. Given the recent incursion of the Ug99 lineage into Iran, there is an increasing likelihood that Ug99 races may have the possibility to interact with *Berberis* spp. occurring widely in CWANA. Barberry species are widely grown in mountainous areas in CWANA including targeted countries in the Ug99 pathway, and their role in epidemiology of wheat rusts is completely unknown. During 2010, stem rust races with high variability for *Sr*-genes were recovered from aecial samples collected from infected Barberry plants in Kelardasht (mountainous areas in Caspian sea region) in Iran indicating that Barberry species can serve as alternate host of Ug99 when it reaches this region. Since greenhouse facilities to work with live samples are not available in many of the countries in this region, it is very important to study the role of barberry as alternate host of wheat stem and yellow rust pathogen with alternative techniques such as molecular diagnostic tools. In the frame of the DRRW project, the Wheat Rust Toolbox was expanded, to store, analyse and display data from this and other DRRW activities dealing with rust on barberry and the role of barberry as alternate host for rust diseases.

Methods

Berberis leaves infected with aecia were collected in the CWANA project in Iran, Azerbaijan, Uzbekistan, Georgia, and Tajikistan. In each country, infected Berberis leaf samples were collected from dispersed areas and from the greatest variety of Berberis species as possible. A special Berberis surveillance and sampling protocol was developed. Dried infected Berberis leaves were sent to the Global Rust Reference Center (GRRC). Single aecial clusters (eq. single lesions) were cut from the Berberis leaves and subjected to DNA-extraction. DNA regions of the translation elongation factor gene (TEF1α), beta-tubuline gene (β-tub) or the Internal Transcribed Spacer (ITS) region were PCR amplified with primers which specifically target the fungal DNA. The PCR products were sequenced and the sequences compared to sequences in GenBank at NCBI and "in house sequences" of *Puccinia* species in order to identify the rust species present on the Berberis leaves.

The Host-Pathogen Database in the Wheat Rust Toolbox was expanded to cover *Berberis* spp. as an alternate host for several *Puccinia* species. A web form for entering the results of sequencing was included as well as maps and tables for display of the results. Results are currently available after login at the Wheat Rust Toolbox and at the GRRC web site.

Results

Host-pathogen database

The wheat Rust Toolbox Host-Pathogen Data Base (DB) is hosting all types of host and pathogen data i.e. wheat rust surveillance data, barberry surveillance and molecular data, pathogen virulence and pathotype data, differentials, differential sets and distribution sets. The restructuring of this DB resulted in a Database structure enabling increased flexibility regarding all possible combinations of host and pathogen information in time and space and from Genus to forma level. Organisms are described by Genus, Species and Forma. An example of the Host-Pathogen DB user interface is given in Figure 1.

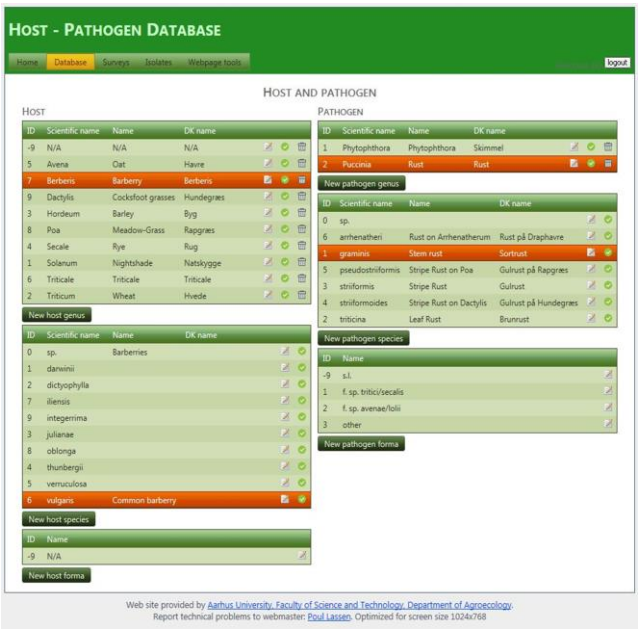


Figure 1. The host is described by host genus name, species name and forma specialis. The Pathogen is described by pathogen genus, species and forma specialis. *Puccinia graminis* is so far defined with three forma specialis, but more can be added:

- *Puccinia graminis f.sp. tritici*
- *Puccinia graminis f.sp. avenae*
- *Puccinia graminis f.sp. secalis*

S.l. is *Sensu latu = miscellaneous, in this context several variants of P. graminis which at present could not be assigned to the tritici, avenae or secalis group.*

Based on the new Host-Pathogen DB it is now possible to handle all kinds of pathogen data i.e. virulence/pathotypes, molecular diagnostics SNP data, biological assays and BLAST results. It is possible to link the results from lab/greenhouse analysis with the wheat rust surveillance data and the original stem rust pathotype data can be affiliated with geo-reference data. The system is prepared for the implementation of a Track and Trace Isolates Information System (TTIIS), - to handle future situations where the same isolate will be analysed with several methods and in different labs, e.g. PCR molecular diagnostics in one lab, DNA sequence analysis in another lab, traditional pathotyping and infection assays in yet another lab. This will need coordination and harmonization of terms, definitions and other agreements between partners.

Two major core types of data are currently implemented and published within the Wheat Rust Toolbox – rust survey data and data about the pathogen isolates sampled. The wheat rust survey database currently holds more than 12,500 geo-referenced, standardized, field survey data on all three wheat rusts and covering 34 countries, from 2007 to 2013. For Barberry we only have data from approximately 150 locations. The database structure enables the storage of survey data with no sampling of isolates, and storage of isolate data information that is not associated with a specific survey. The link between the survey data and the Isolate data via a sample ID table. When isolates have been sampled they might be analyzed with several different methods in different labs in the network (Figure 2).

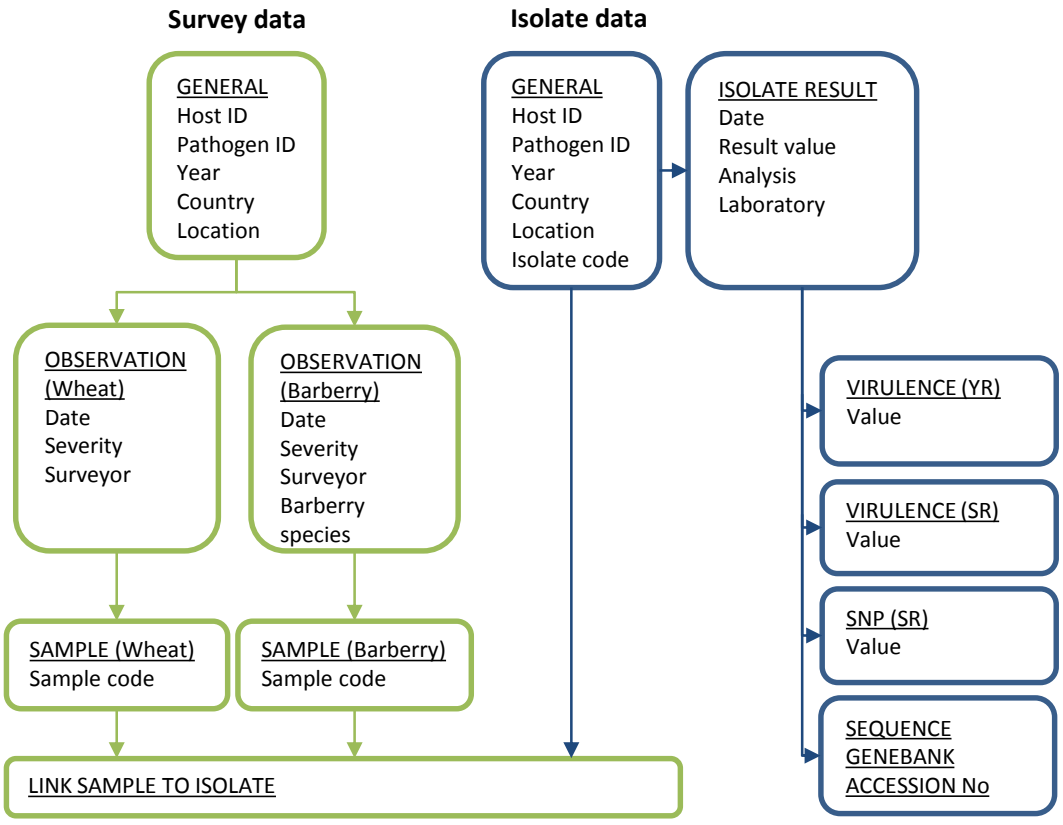


Figure 2. General structure of the Host-Pathogen Database

Rust on Barberry Data Management and Dissemination Tools

Dataflow and system description of the Barberry data management and dissemination tool is shown in Fig. 3. The system is based on a modern ICT three layer approach – separation of the database, the models and application layer and the view/display layer. This enables exchange of data and models between the wheat rust toolbox and other similar information systems, and, that dissemination tools easily can be embedded in partner web sites via iFrame technology (Hodson et al., 2012). The database and web system is designed to cope with specific objectives and sources of data:

General objectives of Barberry surveillance and monitoring

- Where are Barberry found?
- Which type of rust is found on Barberry?
- Which Barberry species are infected?

Sources of data

- Surveillance data organized by ICARDA / CIMMYT. Barberry leaf samples sent to GRRC
- DNA samples tested by GRRC against known DNA sequences in GenBank (NCBI) and in GRRC local gene bank.

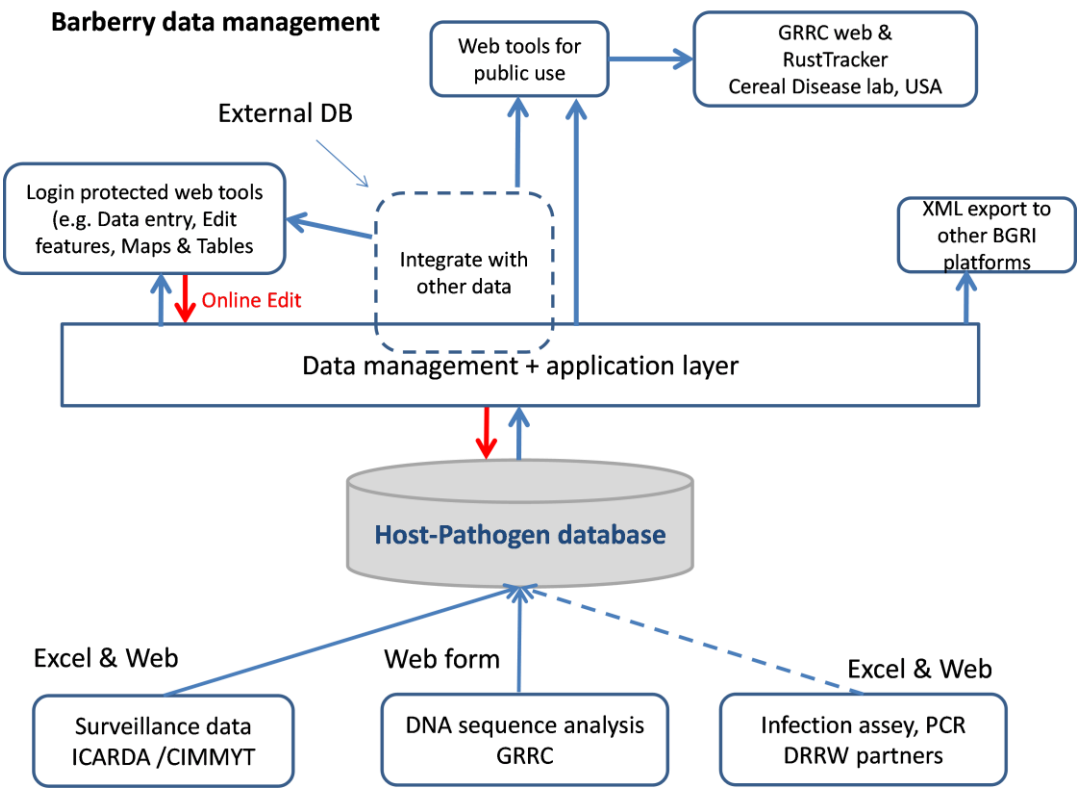


Figure 3. The Barberry management, dataflow, dissemination and ICT framework. The dotted lines indicate the system is prepared for this activity, but not yet implemented

Data entry web form

The "Rust survey on barberry" data entry form (Fig. 4) was implemented based on an Excel version defined for the CWANA project by ICARDA. In the web form the user can only enter results as predefined options and geo-reference data are immediately shown on a map. This ensures high quality and comparable results. The entry of sample ID is the link between the survey data and results from analysis of the pathogen sampled. When data have been uploaded and quality controlled, a data management system is available after login for restricted users to Edit, Delete, and make data public / un-public and for export of data to XML or CSV files (Fig 5).

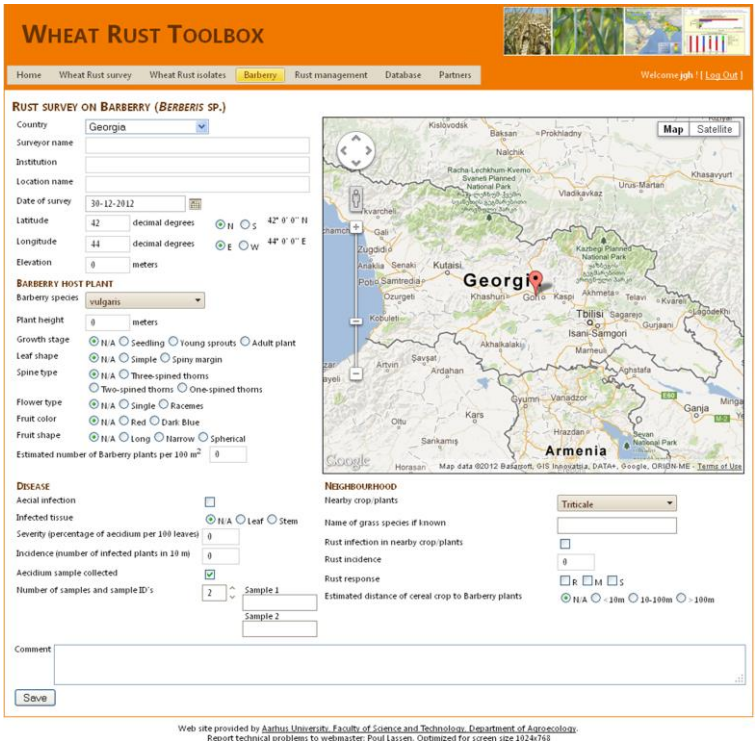


Figure 4. Rust on barberry survey data entry web form. This is available after login to the Wheat Rust Toolbox. The system will know which country you are affiliated with. The Google map will display the location of the geo coordinates entered. The user enters information via drop down selection boxes, radio buttons and checkmark selections.

Using the arrows for Number of samples, it is possible to report several samples collected at the same location. The Sample IDs entered will be available during upload of isolate information i.e. DNA molecular diagnostics results or pathotyping.

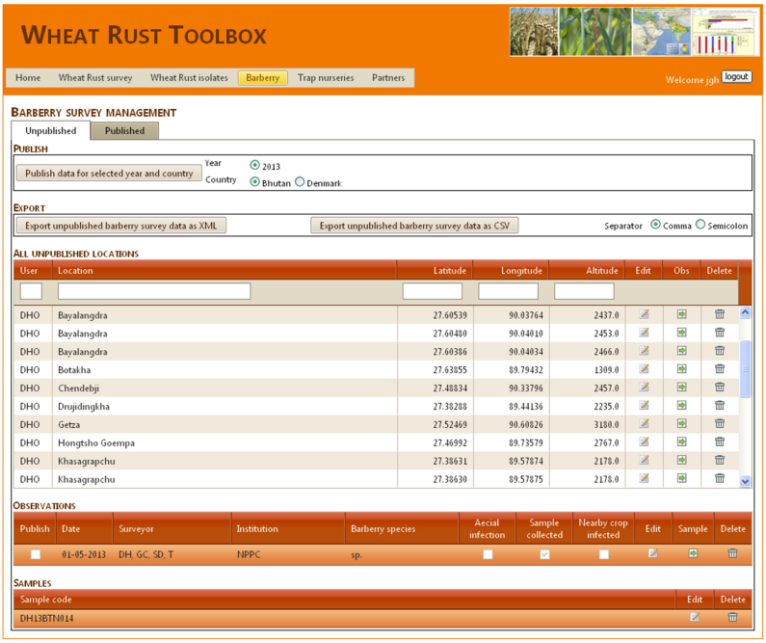


Figure 5. After upload of data a few "Barberry managers" can Publish data for selected years and countries, Export unpublished or published data to XML or CSV, Edit or delete data.

Currently, observations from 133 barberry survey sites from the CWANA project have been imported to the database. Observations are from 2011, 2012 and from the countries Azerbaijan, Georgia, Iran, Tajikistan, Uzbekistan. Most recently 5 observations were entered from a survey in Bhutan (Table 1). Leaf samples with infections from these locations were received at GRRC for DNA sequence analysis.

Table 1. Number of barberry survey sites 2011, 2012 and 2013.

Country Name	2011	2012	2013	Total
Azerbaijan	0	3	0	3
Bhutan	0	0	5	5
Georgia	0	15	0	15
Islamic Republic of Iran	0	60	0	60
Tajikistan	49	0	0	49
Uzbekistan	0	6	0	6
Total	49	84	5	138

Entry of data from DNA sequence analysis

When selecting menu item Barberry/DNA sample/new DNA sample on barberry, initially the user select year, country and sample code from the barberry survey database (Fig. 6). This is how the Lab results are linked with the survey data. After selection, the system will display the basic data from the survey database e.g. location, collection date, surveyor name, institution and if any comments. The rest of the webform is for entering results from the DNA sequence test i.e. DNA sample code, DNA Sequence data. DNA sequence accession number in gene bank to which max identity was found. Currently Genbank (NCBI) and GRRC can be selected and results are reported as Accession number, Direct Link to accession number in GenBank, Description from the Gene bank, Max identity found, Comment and Conclusion of test what species was found. It is possible to enter more than one result from the same gene bank and it will be possible to enter results from more gene banks. Current options are GenBank (NCBI) and GRRC's own bank of reference sequences. Options for *Puccinia* species are sp., *P. arrhenatheri*, *P. graminis*, *pseudostriiformis*, *striiformis* and *striiformoides*. Options for forma are: N/A, *f. sp. tritici, f. sp. avenae f. sp. secalis* and *S.l.* The four options for forma are only available after selecting species=graminis. Currently, for all other species the forma option will be N/A.

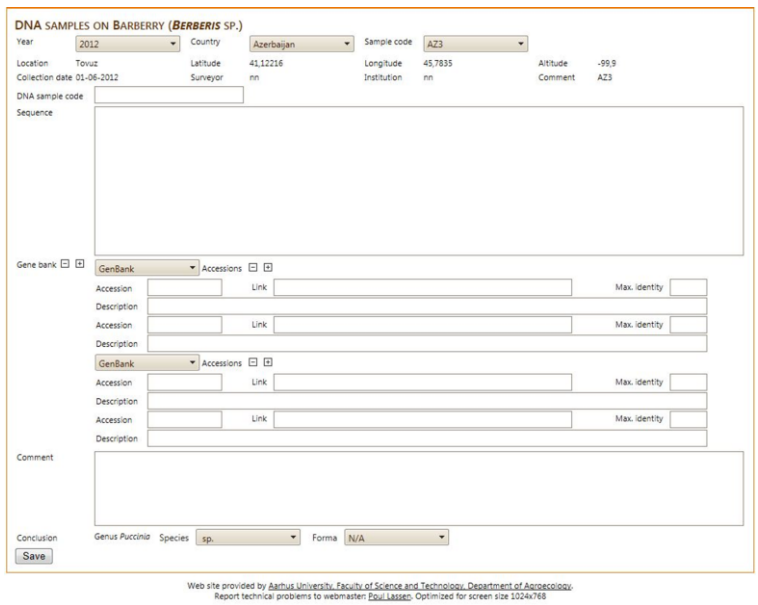


Figure 6. Data entry web form for DNA sequence analysis data. The user can add one or more gene banks via the Gene bank - + buttons. For selected gene bank name the user can add more results lines via the Accessions - + buttons. In the example shown two gene bank names were selected and for each gene bank max identity with two accession numbers are given as a result

See text for further explanation.

Edit tools

After upload of DNA sequence data, they can be edited via the "Edit DNA samples on barberry" web page. In the top table the user selects the Edit button ( ) to edit Code, Pathogen species, forma and comment. For each sample the table for accession results is opened via the Accession link to results button ( ).

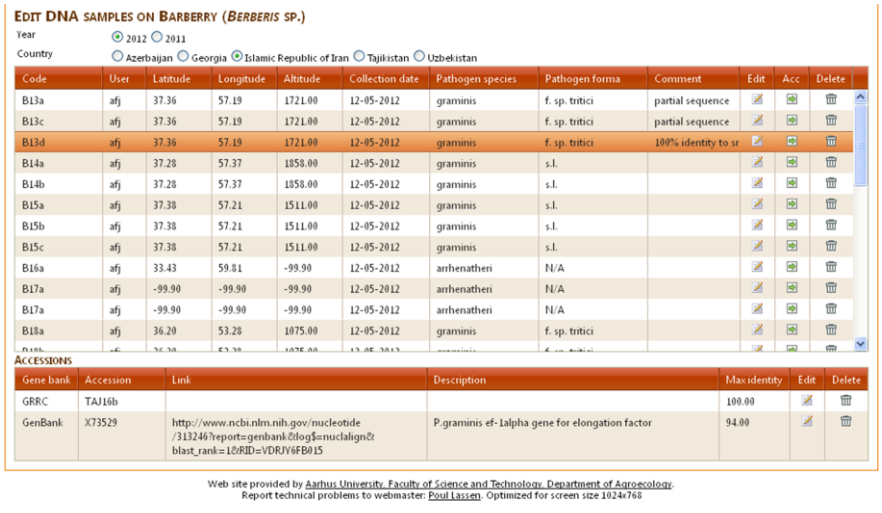


Figure 7. The Edit DNA samples web page. DNA sample code B13D is selected and the green arrow for accession number is also ticked. In the table below the main table, is given Gene bank name, Accession number and Max identity tested against the "in house" GRRC collection and the GenBank (NCBI).

The comment for DNA sample code B13D was: 100% identity to Stem rust from *Hordeum vulgare* sampled in Tajikistan. The max identity is available for editing when selecting the Edit button in the Accessions table (Fig. 8)

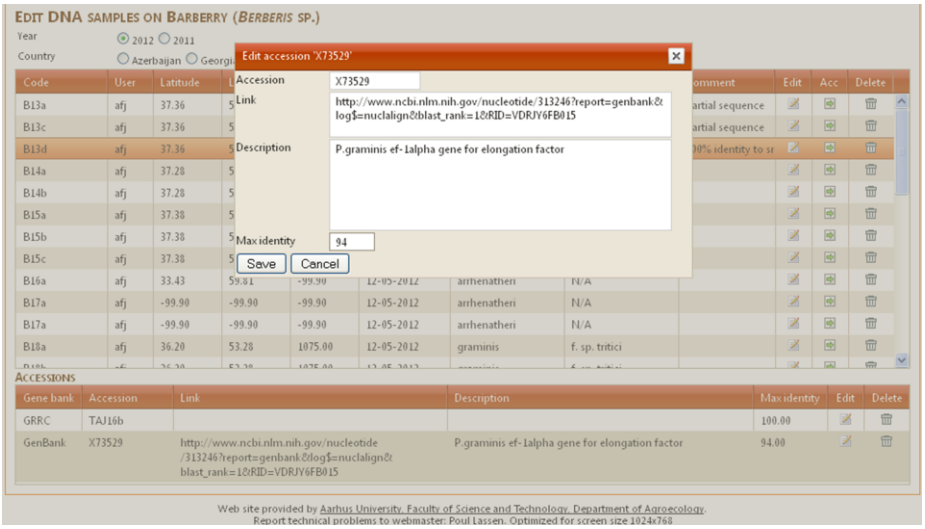


Figure 8. Pop up Edit window for accession X73529.

Display tools

The barberry web tools are divided into data entry web pages, tools for editing and tools for the display of results. Edit tools and the Part of the display tools showing primary data will only be available for restricted user groups (Figures 5, 7, 8 and 11)

The observations of rust on Barberry are shown on a mapping tool similar to the Wheat rust survey mapper. In a first draft version observations are shown as blue and yellow dots, indicating where barberry plants have been inspected for rust aecia, and in most cases sampled for DNA analysis (Fig. 9). Locations with blue color indicate the DNA samples have been analysed successfully and results are available. Click on the dots to see attribute data, results and if available photos (under development). Improvements could be that icons and codes are changed to squares with different colors indicating what type of rust was found on the Barberry plants at the locations displayed. Options to display could be:

- *Puccinia graminis* f. sp. *tritici* (Stem rust)
- *Puccinia graminis* f. sp. *avenae* (Oat stem rust)
- *Puccinia graminis* f. sp. secalis (Rye stem rust)
- *Puccinia graminis* S.l (Stem rust, forma specialis not determined)
- *Puccinia striiformis* (Wheat stripe rust)
- *Puccinia pseudostriiformis* (Poa stripe rust)
- *Puccinia striiformoides* (Dactylis stripe rust)
- *Puccinia arrhenatheri* (Arrhenatheri rust)
- Unknown

More species can be added to the list when necessary. By selecting a square on the map a pop-up menu will display attribute data including results from lab test, i.e. Lab, method and result. For the results that currently are being uploaded to the DB by GRRC, this will be in the form:

- Lab: GRRC
- Method: DNA sequence analysis
- Result: *Puccinia graminis*. S.l.

It should be possible to checkmark one or more *Puccinia* species / forma specialis (f.sp.) enabling search for the regional distribution of one or more specific species. Several species may be found at the same location and this feature will overcome the problem that only one icon from a certain location will be visible on the map.

Verified results from the DNA analysis by country and year are summarized in the “DNA samples Map” (Fig. 10). The user can select one or more years and results are given as frequencies of different *Puccinia* species/f.sp. found on Barberry in a country that year.

A summary of all results is available in a sortable table for a restricted “Barbery managers” group after login at the toolbox (Fig. 11)

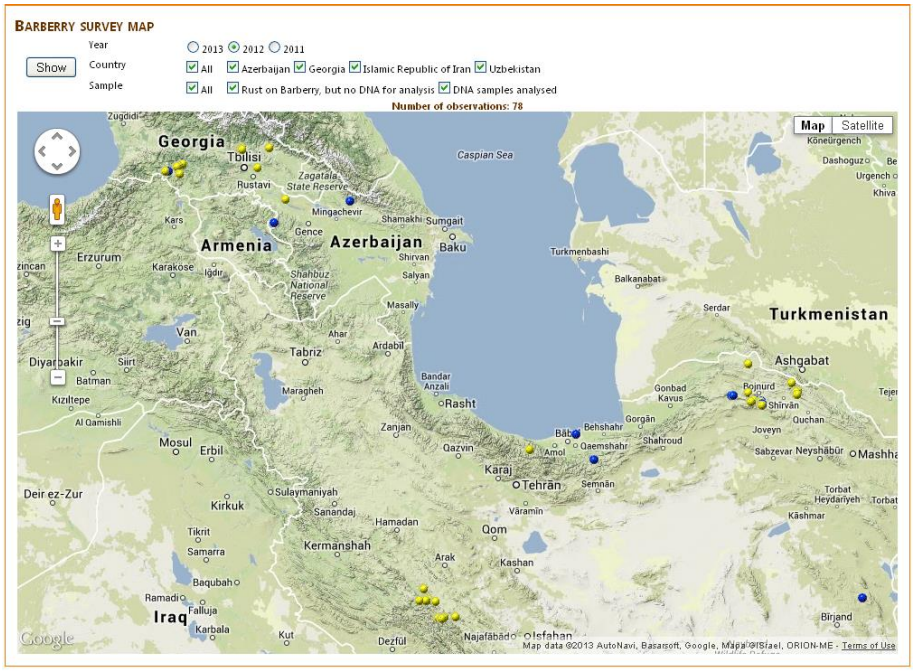
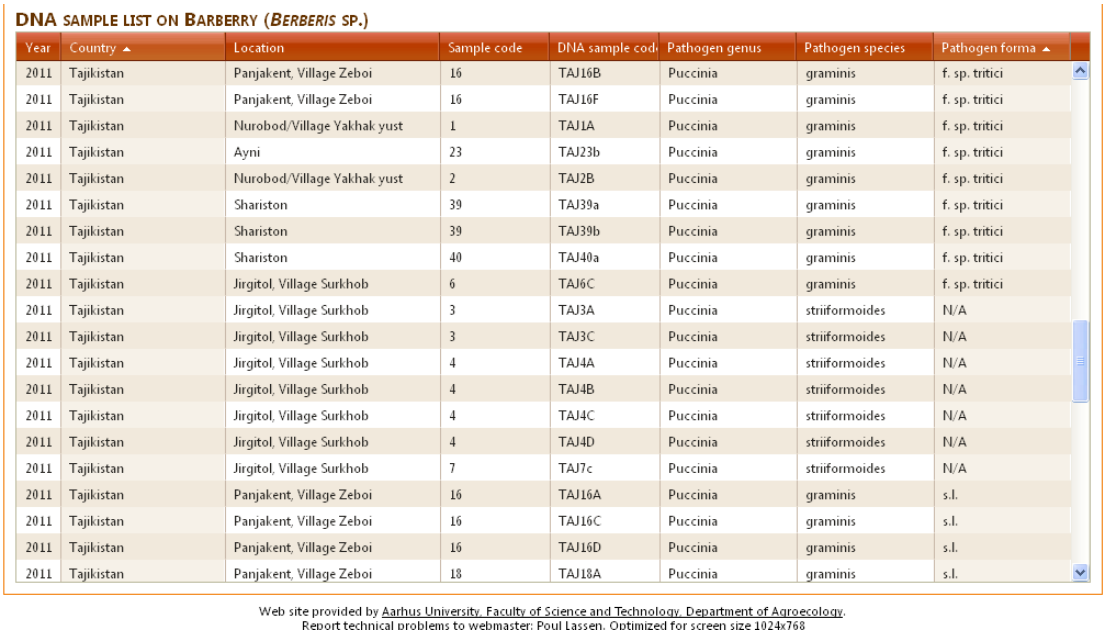


Figure 9. Rust on Barberry mapper showing where rust on Barberry has been found in 2012 (selected). Blue dot: DNA analysed successfully. Yellow dot: leaf sample not received by GRRC (only for Georgia), or, Isolation of DNA failed, or, analysis not yet completed.

Figure 10. Screen dump of the "DNA samples Map" display Web tool. The user can select one or more years. Data are given as frequencies of different *Puccinia* species found on Barberry in a country that year, or combination of years. Country code and number of samples are displayed below the pie charts.



DNA SAMPLE LIST ON BARBERRY (*BERBERIS* SP.)

Year	Country	Location	Sample code	DNA sample cod	Pathogen genus	Pathogen species	Pathogen forma
2011	Tajikistan	Panjakent, Village Zeboi	16	TAJ16B	Puccinia	graminis	f. sp. tritici
2011	Tajikistan	Panjakent, Village Zeboi	16	TAJ16F	Puccinia	graminis	f. sp. tritici
2011	Tajikistan	Nurobod/Village Yakhak yust	1	TAJ1A	Puccinia	graminis	f. sp. tritici
2011	Tajikistan	Ayni	23	TAJ23b	Puccinia	graminis	f. sp. tritici
2011	Tajikistan	Nurobod/Village Yakhak yust	2	TAJ2B	Puccinia	graminis	f. sp. tritici
2011	Tajikistan	Shariston	39	TAJ39a	Puccinia	graminis	f. sp. tritici
2011	Tajikistan	Shariston	39	TAJ39b	Puccinia	graminis	f. sp. tritici
2011	Tajikistan	Shariston	40	TAJ40a	Puccinia	graminis	f. sp. tritici
2011	Tajikistan	Jirgitol, Village Surkhob	6	TAJ6C	Puccinia	graminis	f. sp. tritici
2011	Tajikistan	Jirgitol, Village Surkhob	3	TAJ3A	Puccinia	striiformoides	N/A
2011	Tajikistan	Jirgitol, Village Surkhob	3	TAJ3C	Puccinia	striiformoides	N/A
2011	Tajikistan	Jirgitol, Village Surkhob	4	TAJ4A	Puccinia	striiformoides	N/A
2011	Tajikistan	Jirgitol, Village Surkhob	4	TAJ4B	Puccinia	striiformoides	N/A
2011	Tajikistan	Jirgitol, Village Surkhob	4	TAJ4C	Puccinia	striiformoides	N/A
2011	Tajikistan	Jirgitol, Village Surkhob	4	TAJ4D	Puccinia	striiformoides	N/A
2011	Tajikistan	Jirgitol, Village Surkhob	7	TAJ7c	Puccinia	striiformoides	N/A
2011	Tajikistan	Panjakent, Village Zeboi	16	TAJ16A	Puccinia	graminis	s.l.
2011	Tajikistan	Panjakent, Village Zeboi	16	TAJ16C	Puccinia	graminis	s.l.
2011	Tajikistan	Panjakent, Village Zeboi	16	TAJ16D	Puccinia	graminis	s.l.
2011	Tajikistan	Panjakent, Village Zeboi	18	TAJ18A	Puccinia	graminis	s.l.

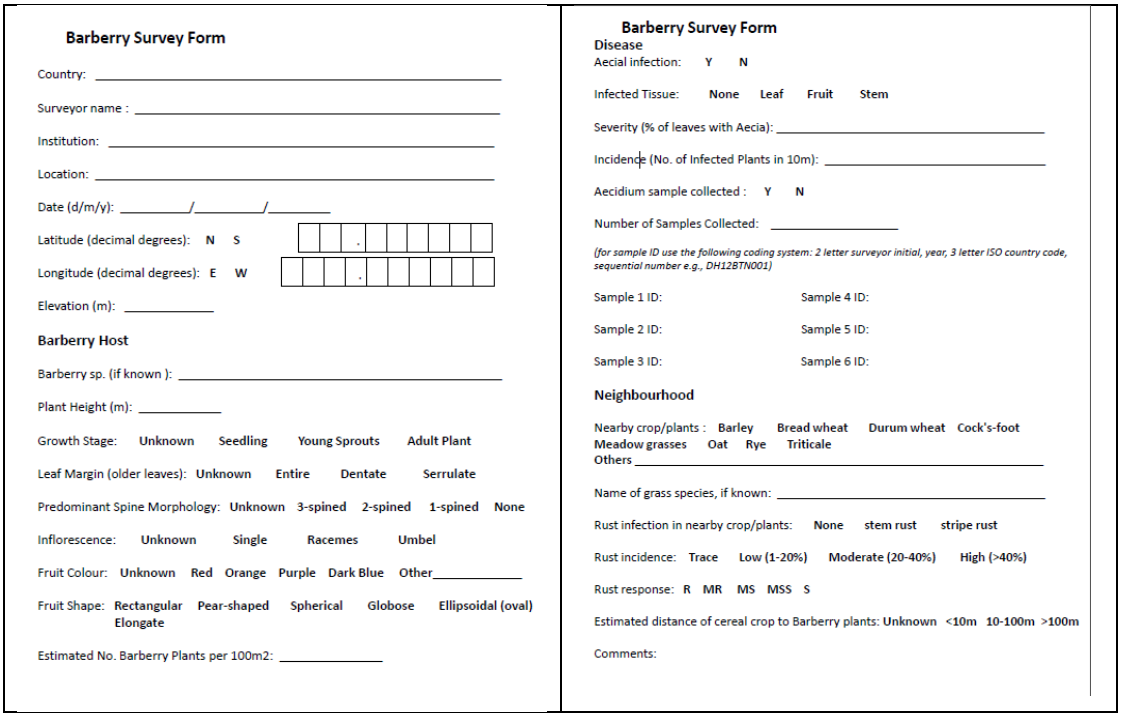
Web site provided by Aarhus University, Faculty of Science and Technology, Department of Agroecology.
Report technical problems to webmaster: Poul Lassen. Optimized for screen size 1024x768

Figure 11. Screen dump of the "DNA sample list on Barberry" display Web tool. The table can be sorted by descending or ascending by selecting up to three header titles. In this example the table is sorted according to Country and then Pathogen forma.

Discussion points

Barberry survey form

- A modification of the Barberry survey form was proposed by David Hodson et al. in May 2013 (Fig. 12). This form was elaborated and used during a survey in Bhutan, May 2013. During the "Berberis rust survey in the Ug99 pathway in CWANA" project, only one partner delivered all attribute data. All other partners just delivered plant samples, location names and geo-reference data. It seems to be a barrier for this type of survey to collect all background information. It might be possible to collect all attribute data during the DRRW project when surveyors are paid by the project, but the questions is if this will be possible in the long run when this support ends. A final survey form should be discussed and how to train and motivate surveyors to sample all necessary data with high quality and in time.
- Several questions are related to the taxonomy of the Berberis plants. This might be a barrier for non-experts. One option to improve on this is to develop a field protocol with identification material including photos and drawings as well as web based material for taxonomical identifications, training material etc.
- For future surveys one should ask: What questions are the most important to be answered. Who are the target audience for this information? What kind of information and tools can be used to answer the questions and finally what data are needed for the tools that can answer the questions.
- Since the time and location of barberries survey is normally different from wheat growing areas, separate survey programs and funds are required for Berberis surveys.



Barberry Survey Form

Country: _____

Surveyor name : _____

Institution: _____

Location: _____

Date (d/m/y): _____/_____/_____

Latitude (decimal degrees): N S

Longitude (decimal degrees): E W

Elevation (m): _____

Barberry Host

Barberry sp. (if known): _____

Plant Height (m): _____

Growth Stage: Unknown Seedling Young Sprouts Adult Plant

Leaf Margin (older leaves): Unknown Entire Dentate Serrulate

Predominant Spine Morphology: Unknown 3-spined 2-spined 1-spined None

Inflorescence: Unknown Single Racemes Umbel

Fruit Colour: Unknown Red Orange Purple Dark Blue Other_____

Fruit Shape: Rectangular Pear-shaped Spherical Globose Ellipsoidal (oval) Elongate

Estimated No. Barberry Plants per 100m2: _____

Barberry Survey Form

Disease

Aecial infection: Y N

Infected Tissue: None Leaf Fruit Stem

Severity (% of leaves with Aecia): _____

Incidence (No. of Infected Plants in 10m): _____

Aecidium sample collected : Y N

Number of Samples Collected: _____

(for sample ID use the following coding system: 2 letter surveyor initial, year, 3 letter ISO country code, sequential number e.g., DH12BTN001)

Sample 1 ID: Sample 4 ID:

Sample 2 ID: Sample 5 ID:

Sample 3 ID: Sample 6 ID:

Neighbourhood

Nearby crop/plants : Barley Bread wheat Durum wheat Cock's-foot Meadow grasses Oat Rye Triticale

Others _____

Name of grass species, if known: _____

Rust infection in nearby crop/plants: None stem rust stripe rust

Rust incidence: Trace Low (1-20%) Moderate (20-40%) High (>40%)

Rust response: R MR MS MSS S

Estimated distance of cereal crop to Barberry plants: Unknown <10m 10-100m >100m

Comments:

Figure 12. Barberry survey form used for the rust on barberry survey in Bhutan, 2013.

Sampling strategy

- For some samples from the CWANA project it was not possible to extract DNA for the sequence analysis. How to optimize sampling time and method should be discussed and a common method should be developed.
- How will it be possible to apply different methods to the same samples (molecular diagnostics, infection assay etc. carried out in the same or in different labs). The infection assay need live samples, but other methods can rely on dead samples.
- A major part of the results in this study indicate *Puccinia graminis* S.l. (sensu lato = in a relatively broad sense). This means that it is identified as *P. graminis.* Currently, it is not possible to identify forma specialis, based on the molecular diagnostic method used in the CWANA project. In areas with high genotype diversity of wheat rust we should consider to sample rust on different host species neighboring the barberry bushes to enable, - not only analyzing DNA via comparison to DNA sequence in GenBank, but also to DNA sequences from rust sampled on wheat and wild grasses in the local area. This can be included in a sampling strategy.

Synergies and added values via collaboration

- We should discuss how to obtain comparable results to obtain robust conclusion in a global or regional context i.e. how to upload data on infection assay and PCR diagnostics data and similar data to the Wheat Rust Toolbox to be analyzed and displayed in a common and integrated framework
- We offer to collate and store all results regarding rust on barberry from the DRRW project and related projects in the Wheat Rust Toolbox
- Partners will have access to own results. No primary data will be delivered to any third party.

Dissemination of results

- In the Wheat Rust Toolbox summary data as maps and graphics will be developed as "show versions" that easily can be embedded in any web site (see RustTracker.org and wheatrust.org). The same information can be integrated in several websites with different context.

Participants

CIMMYT
David Hodson, Senior Scientist at CIMMYT, Ethiopia, main responsible for the BGRI Global Rust Monitoring and Surveillance System / RustTracker

Aarhus University
Jens Grønbech Hansen and **Poul Lassen**, Department of AgroEcology are responsible for the development of the Wheat Rust Toolbox databases and associated tools.

Annemarie F. Justesen, Department of AgroEcology main working area is molecular plant pathology with special focus on molecular diagnostics and population biology. She is part of the Global Rust Reference Centre at Research Centre Flakkebjerg. She did all the molecular lab work and sequencing

Mogens S. Hovmøller, Department of AgroEcology is leader of the Global Rust Reference Center

ICARDA
Kumarse Nazari, Senior Cereal Pathologist, ICARDA, is rust pathologist, regional coordinator of BGRI surveillance in CWANA and was responsible for coordinate/ surveys and sampling in the current project.

Literature

Hodson DP, Hansen JG, Lassen P, Alemayehu Y, Arista J, Sonder K, Kosina P, Moncada P, Nazari K, Park RF, Pretorius ZA, Szabo LJ, Fetch T & Jin Y (2012) Tracking the Wheat Rust Pathogen. In: *Proceedings Borlaug Global Rust Initiative 2012 Technical Workshop: Oral Presentations.* McIntosh, R. (red.). bgri@cornell.edu

Hovmøller MS, Sørensen CK, Walter S, and Justesen AF (2011). Diversity of Puccinia striiformis on Cereals and Grasses. *Annual Review of Phytopathology* Vol. 49: 197-217 DOI: 10.1146/annurev-phyto-072910-095230

Hovmøller M S,; Justesen AF, Brown JKM. (2002) Clonality and long-distance migration of *Puccinia striiformis f.sp. tritici* in north-west Europe. *Plant Pathology*, Vol. 51, Nr. 1, p. 24-32.

Jin Y, Szabo L, Carson M (2010). Century-old mystery of Puccinia striiformis life history solved with the identification of *Berberis* spp. as an alternate host. *Phytopathology* 100:432–435

Jin Y (2011). Role of Berberis spp. as alternate hosts in generating new races of *Puccinia graminis* and *P. striiformis. Euphytica (2011) 179:105–108*
DOI 10.1007/s10681-010-0328-3

Mboup M, Leconte M, et *al*. (*2009*). Evidence of genetic recombination in wheat yellow rust populations of a Chinese oversummering area. *Fungal Genet. Biol.*

Duan X, Tellier A, Wan A, Leconte M, de Vallavieille-Pope C, Enjalbert J (2010) Puccinia striiformis f.sp.tritici presents high diversity and recombination in the over-summering zone of Gansu, China. *Mycologia, 102(1), 2010, pp. 44–53.*
DOI: 10.3852/08-098

Zhao J, Wang L, Wang Z, Chen X, Zhang H, Yao J, Zhan G, Chen W, Huang L, Kang Z. (2013). Identification of Eighteen Berberis Species as Alternate Hosts of *Puccinia striiformis f. sp. tritici* and Virulence Variation in the Pathogen Isolates from Natural Infection of Barberry Plants in China. *Phytopathology. 2013 Mar 20*

Wellings CR (2011) Global status of stripe rust: a review of historical and current threats. *Euphytica* May 2011, Volume 179, Issue 1, pp 129-141