INTERNATIONAL WORKSHOP

SURVEILLANCE AND CONTROL OF CASSAVA DISEASES IN AFRICA

PÔLE DE PROTECTION DES PLANTES (3P) SAINT-PIERRE, LA RÉUNION ISLAND

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DIAGNOSTIC TOOLS AT THE 3P CENTER

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team at the 3P Center is developing diagnostic molecular tools for quarantine bacterial and viral plant diseases. The work relies on the abundant and various data obtained from taxonomy, diversity, epidemiology, and genomic studies of the different pathogens, generated by the 3P Center team and collaborators.

Several sensitive and specific PCR-based protocols have been developed to detect bacterial and viral pathogens (e.g detection of Xanthomonas axonopodis pv. dieffenbachiae in Anthurium tissues by nested PCR, (Robène-Soustrade et al., 2006. AEM 72: 1072-1078), detection of X. axonopodis pv. allii in onion seeds by duplex-nested PCR (Robène-Soustrade et al., 2010. AEM 76: 2697-2703), detection and quantification of a wide range of begomoviruses by five duplex real-time quantitative PCRs (Péréfarres et al., 2011. Virol J 8: 389). These methods are useful diagnostic tools for indexing propagative plant material and for international sanitary surveillance of plant material exchanges. These protocols are intended to be referenced as French official methods and EPPO standards (e.g. X. axonopodis pv. dieffenbachiae). The team has an expertise in comparing and validating different molecular tools (Delcourt et al., 2013. Plant Dis 97: 373–378) and collaborates with the French agency for food, environmental and occupational health safety (ANSES) to validate the different protocols through both intra laboratories and ring tests involving different European laboratories (Chabirand et al., 2014. Plant Pathol 63: 20–30). We are also innovating into new DNAbased diagnostic technologies by developing an efficient and portable microarray technology to detect and identify different pathogenic and/or genetic groups of *Ralstonia solanacearum*.

The team's competencies span development, assessment, and validation of diagnostic tools as well as management of collaborative studies. We can rapidly adapt to new challenges such as the evaluation and improvement of the sanitary situation of cassava in Africa. We propose to assess different existing diagnostic tests and to innovate into new DNA-based diagnostic technologies if necessary, in order to optimize the diagnostic of the main viral and bacterial pathogens of cassava in Africa.

BROAD RANGE VIRUS INDEXING THROUGH NGS: THE SAFE-PGR CASE STUDY.

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B iological Resources Centers (BRCs) conserve and distribute plant germplasm for research and development purposes. As such, they play a strategic role by providing breeding programs with genitors that are critical for crop adaptation to ongoing environmental and socie-

tal changes. BRCs must guarantee the sanitary status of the resources they distribute, in order to prevent the spread of diseases, particularly for vegetative propagated that do not benefit from the virus sanitation occurring through a seed cycle. The Safe-PGR project (*Towards Safer Plant Genetic Resources through improved viral diagnostics*) was initiated in 2012 to improve the knowledge of the viruses infecting the crops addressed by four partner's BRCs in Guadeloupe, Madeira, Azores and Reunion, and develop classical or new diagnostic techniques for the species they deal with: banana, garlic, sugarcane, sweet potato, vanilla and yam. The project is funded by the French National Agency for Research and the governing bodies of Azores, Madeira, Guadeloupe and Reunion. The research consortium involves teams from INRA (BFP, ASTRO), CIRAD (BGPI, AGAP, PVBMT), CBA Azores and ISOPLEXIS Madeira.

Methods: The project aims at exploring the molecular diversity of the viral families affecting the targeted crops, optimize classical diagnostic methods taking into consideration data generated through this analysis of viral diversity and develop new multi-pathogen diagnostic methods based on metagenomics and deep-sequencing technologies (Roche 454). Eight nucleic acids extraction methods for metagenomics studies have been tested and compared. Two complementary methods based on the extraction of double-strand RNA and viral particles have been selected. Bioinformatics tools have been successfully developed for analyzing the metagenomics data. These methods are currently used for the screening of 1500 plants from the CRB germplasm collections.

Virus discovery : The preliminary bioinformatics analyses of plant EST databases and of the first deep sequencing results generated, allowed the tentative identification of a total of 25 new viruses in Garlic, Sugarcane, Yam and Vanilla for which new and efficient detection assays have been developed and implemented. Further characterization of a new *Allexivirus* of garlic and new *Potexvirus* of vanilla will be presented.

IT CAPACITIES IN LA RÉUNION ISLAND



a Réunion island The Plant Protection Platform Center (3P Center) is CIRAD's largest laboratory and hosts and manages most of the technical IT resources for the entire island.

Henri BROUCHOUD IT Manager, Cirad, France

The IT infrastructure includes all that is necessary for high level science activities : virtualized datacenter, high bandwidth networks, high performance workstations...

Since 2003 the 3P Center has been involved in several regional programs and projects in which it has taken the lead in IT activities, especially the creation and development of a Web portal for biodiversity and sustainable agricultural production. The 3P Center has also helped to develop a database of regional pests and diseases that contains a visual inventory of plant pests and diseases in the Indian Ocean area.

Another project involves innovative applications for smartphones that can automatically recognize plants from pictures, or observation statements and visual diagnosis of plant diseases on the spot.